

*An update to the fatty acid profiles of bovine retail milk in the United Kingdom: implications for nutrition in different age and gender groups*

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**An update to the fatty acid profiles of bovine retail milk in the United Kingdom:  
implications for nutrition in different age and gender groups**

**Running title: Fatty acid profile of UK retail milk and dietary intake implications**

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## Abstract

This study investigated the effect of UK dairy production system, month, and their interaction, on retail milk fatty acid (FA) profile throughout the year. Milk samples (n=120) from four conventional (CON), four organic (ORG) and two free-range (FR) brands were collected monthly. ORG milk had more nutritionally-desirable polyunsaturated FA, including rumenic acid and the omega-3 PUFA  $\alpha$ -linolenic, eicosapentaenoic and docosapentaenoic acids, and less of the nutritionally-undesirable palmitic acid. Milk FA profile was similar between FR and CON, but FR milk had less SFA and/or palmitic acid, and/or greater  $\alpha$ -linolenic and rumenic acids in certain months within the peak-grazing season. According to the measured milk FA profiles and UK milk fat intakes, milk and dairy products contribute around one-third of the maximum recommended saturated FA intake. A small increased intake of beneficial PUFA may be expected by consuming ORG milk but human health implications from such differences are unknown.

**Keywords:** milk; dairy management; dietary intakes; fatty acids; free-range; human health; omega-3; organic

## 1. Introduction

Milk and dairy products provide a range of beneficial nutrients for human health, including fatty acids (FA), proteins, bioactive peptides, minerals, carotenoids and vitamins (Haug, Hostmark, & Harstad, 2007; Pereira, 2014; Thorning, et al., 2017). However, milk and dairy products are dietary sources of saturated fatty acids (SFA), such as C12:0, C14:0 and C16:0, elevated consumption of which may increase the risk of cardiovascular disease (CVD) (EFSA, 2010; FAO, 2010). These concerns and the increased incidence of lifestyle-related diseases, such as obesity and CVD, may have contributed to the reduction in whole milk consumption in developed countries, including UK, Denmark, France, USA, Canada and Germany (Kliem & Givens, 2011). In the UK, whole milk consumption has decreased 5-fold compared with 1970s' levels, and despite the simultaneous increase in semi-skimmed milk consumption, the overall milk intake has declined (Kliem, et al., 2011). In contrast, milk is also rich in FA with potentially beneficial effects on human health (see reviews from (Barcelo-Goblijn & Murphy, 2009; Dilzer & Park, 2012; Field, Blewett, Proctor, & Vine, 2009; Haug, et al., 2007; Swanson, Block, & Mousa, 2012)), such as the monounsaturated FA (MUFA) t11 C18:1 (VA, vaccenic acid) and c9 C18:1 (OA, oleic acid), the polyunsaturated FA (PUFA) c9c12c15 C18:3 (ALNA,  $\alpha$ -linolenic acid), c5c8c11c14c17 C20:5 (eicosapentaenoic, EPA), c7c10c13c16c19 C22:5 (docosapentaenoic, DPA) and c4c7c10c13c16c19 C22:6 (docosahexaenoic acid, DHA), which are omega-3 PUFA (n-3), the c9c12 C18:2 (LA, linoleic acid), which is an omega-6 PUFA (n-6), and the conjugated FA c9t11 C18:2 (RA, rumenic acid) (Kliem & Shingfield, 2016; Pereira, 2014).

Current nutritional recommendations are to reduce SFA consumption (as low as possible and not exceeding 10% of total energy intake) and substitute dietary SFA with MUFA and/or PUFA (EFSA, 2010; FAO, 2010). Previous research has shown that dairy management, and especially cow diet, influence milk FA profiles; for example, cows with increased fresh grass intake,

higher dietary forage:concentrate ratio, and/or diets supplemented with plant oils, oilseeds or protected lipids may produce milk with a FA profile that contains less SFA and more n-3 and RA (Chilliard, Glasser, Ferlay, Bernard, Rouel, & Doreau, 2007; Elgersma, 2015; Kliem, et al., 2016). Therefore, potential differences between different dairy production systems, which involve differences in cow nutrition, may reflect on milk FA composition. In the UK, organic milk contained greater concentrations of ALNA, EPA and n-3 PUFA all year round, and less SFA in milk fat, including C16:0, during summer, when compared with conventional milk (Butler, Stergiadis, Seal, Eyre, & Leifert, 2011b; Stergiadis, et al., 2012). A seasonal effect on milk FA composition has been previously demonstrated in UK retail milk (Kliem, Shingfield, Livingstone, & Givens, 2013), which also influences the extent of the compositional differences between organic and conventional milk (Butler, et al., 2011b). However, the interaction between production system and season has been assessed only during January and July (Butler, et al., 2011b), which are potentially among the months with the highest difference in pasture intake in UK dairy systems (Stergiadis, et al., 2012), so a more detailed assessment throughout the year is required.

Fresh grass intake strongly influences n-3 PUFA content of milk fat, as recently highlighted in several multivariate redundancy analyses (Stergiadis, et al., 2015a; Stergiadis, et al., 2015b; Stergiadis, et al., 2012). Bulk tank milk from conventional extensive pasture-based farms, (pasture intake contributing more than 90% of cow dry matter intake), contained more of the potentially nutritionally beneficial, when replacing SFA in human diets, MUFA and/or PUFA and less SFA when compared with conventional and/or organic milk, although differences were not consistent throughout the year or in all studies (Butler, et al., 2008; Stergiadis, et al., 2015b). Recently, free-range milk, certified on farms where cows have access to pasture for a minimum of 180 days/year and being outdoors for a minimum of 23 hours/day during the grazing season, reached the UK market. In the Netherlands, retail milk from dairy farms under a similar

certification scheme, but with less mandatory access to pasture (minimum 120 days/year at pasture and 6 hours/day), had a similar FA profile to retail conventional milk (Capuano, Gravink, Boerrigter-Eenling, & van Ruth, 2015) but potential differences under the UK dairy management practices have not yet been investigated.

This study therefore aimed to (i) investigate the effect of production system (conventional, organic and, for the first time in the UK, free-range), month (March through to February) and their interaction, on retail milk FA profile throughout the year, and (ii) assess the potential implications on the intakes of FA which are relevant to human health.

## **2. Materials and methods**

### *2.1 Experiment/survey design*

All milk samples (n=120) in the present study were collected from retail outlets in England. The survey lasted for 12 months and samples were collected monthly between March 2016 and February 2017. Four brands of conventional milk and four brands of organic milk were sampled monthly from four retail outlets within a 8 km radius of the University of Reading. The only two brands of free-range-certified milk available to UK consumers during the period of this study were obtained monthly from dairies in Lancashire and Gloucestershire. All retail milk samples were whole, pasteurized and homogenized, while conventional and free-range milk had also their fat content standardized to approximately 3.5 and 3.7 g/100g milk, respectively. Milk samples were collected to represent the latest “best before” date, available at the day of sampling, to ensure minimum storage time at retail outlet. Milk samples in commercial packaging were immediately transferred to the laboratories of the University of Reading, and aliquoted into 30-ml sterile polypropylene screw-top containers and were frozen at -20°C until analysis.

### *2.2 Milk analysis*

Concentrations of fat, protein, casein, and lactose were analysed using a Milkoscan FT6000 (Foss Electric, Hillerod, Denmark), while somatic cell count (SCC) was analysed by a Fossomatic (Foss Electric, Hillerod, Denmark), in the National Milk Laboratories (Wolverhampton, UK). Milk FA profiles were analysed by GC flame ionisation detection (Bruker 350 GC, Bruker, Germany) according to previously described methods of esterification and methylation (Chilliard, Martin, Rouel, & Doreau, 2009), and techniques of peak identification and quantification (Kliem, et al., 2013). A combined correction factor, to account for carbon deficiency in the response of flame ionization detector for FA methyl esters with 4-10 atoms of carbon was used (Ulberth, Gabernig, & Schrammel, 1999).

### 2.3 Statistical analysis

Analysis of variance (ANOVA), derived from linear mixed effects models (residual maximum likelihood analysis; REML) (Gilmour, Thompson, & Cullis, 1995) in GenStat (VSN International, 17<sup>th</sup> Edition, Hempstead, UK), by considering management (Conventional, CON; Organic, ORG; Free-Range, FR) and month (March, April, May, June, July, August, September, October, November, December, January, February), and their interaction, as fixed factors and milk ID (which was unique for each combination of brand/retailer and management) as a random factor. Significant effect of the main treatments was declared when  $P < 0.05$  and tendencies were declared when  $0.05 < P < 0.10$ . The residual diagnostics of the final model were assessed using normality plots, with no data showing deviation from normality except for SCC which were log-transformed prior to ANOVA. Pairwise comparisons of means ( $P < 0.05$ ) were performed using Fisher's Least Significant Difference test. Milk FA profiles are reported as g/kg milk fat. Atherogenicity index (AI), thrombogenicity index (TI), as markers to indicate potential risk of CVD, were calculated according to Srednicka-Tober et al. (2016), as follows:

- $AI = (C12:0 + 4 \times C14:0 + C16:0) / (MUFA + PUFA),$



•  $TI = (C14:0 + C16:0 + C18:0) / [(0.5 \times MUFA) + (0.5 \times n-6) + (3 \times n-3) + (n-3/n-6)]$ .  
 $\Delta^9$ -desaturase activity index ( $\Delta^9I$ ) was calculated according to Kay et al. (2004) as:

$$\Delta^9I = (c9\ C14:1 + c9\ C16:1 + OA + RA) / (c9\ C14:1 + c9\ C16:1 + OA + RA + C14:0 + C16:0 + C18:0 + VA)$$

For the purposes of the intake calculations, this study assumes that all dairy products produced in the UK have the same FA profile as the whole milk analysed. Intakes of individual FA or FA groups, for males/females/all for the age groups of 4-10/11-18/19-64/65+ were estimated separately as:

FA intake (g/d) = fat intake (g/d) (Bates, et al., 2014)  $\times$  contribution of fat from milk and dairy products (% of total fat intake) (Bates, et al., 2014)  $\times$  0.933 (correction factor representing % of FA in total milk fat) (Kliem, et al., 2013)  $\times$  milk FA concentration (% of total FA).

### 3. Results

All differences discussed in the Results section were statistically significant ( $P < 0.05$ ) unless otherwise stated.

#### 3.1 Milk basic composition

##### 3.1.1 Effect of production system

Significant effect of production system was identified for milk concentrations of fat and lactose (Table 1). Compared with CON and FR milk, respectively, ORG milk contained more fat and less lactose (Table 1). There were no significant differences in milk composition between CON and FR milk (Table 1).

##### 3.1.2 Effect of month

Significant effects of month were identified for milk concentrations of all basic composition parameters (Table 2). Milk contained less fat during May-September and December than in

March-April, with the remaining months showing intermediate values, mostly without being significantly different (Table 2). Protein concentrations in milk were higher in May, October and November (highest) than in June-September, December and February, with the remaining months showing intermediate values, mostly without being significantly different (Table 2). Casein concentrations in milk were highest in October-November when compared with all other months, although the difference with May was not statistically significant (Table 2). Highest lactose concentrations were observed in March-May and lowest in July, September, October and December with intermediate values being observed during the other months, mostly without being significantly different (Table 2). Milk had higher SCC during winter (December-February) than in March-November, although the difference with January was not statistically significant (Table 2).

### *3.1.3 Effect of the production system $\times$ month interaction*

Significant effects of the production system  $\times$  month interaction were identified for milk lactose concentrations (Appendix; Figure A1); CON milk had more lactose than ORG and FR milk in June, September and November, and less lactose than FR milk in July.

## *3.2 Milk FA profile*

### *3.2.1 Effect of production system*

Significant effect of the production system was identified for milk concentrations of C16:0, RA, ALNA, EPA, DPA, PUFA, n-3, and *trans* FA and the ratios of n-3/n-6, TI, C14:1/C14:0, C16:1/C16:0 and OA/C18:0 (Table 1). Compared with CON and FR milk, respectively, ORG milk had lower concentrations of C16:0 and higher concentrations of RA, ALNA, EPA, DPA, *trans* MUFA, PUFA, cis PUFA, trans PUFA, cis/trans plus trans/cis PUFA, n-3 and *trans* FA (Table 1). ORG milk had a higher ratio of n-3/n-6 and lower ratio of TI, C14:1/C14:0, C16:1/C16:0 and OA/C18:0 than CON and FR milk although the difference between ORG and

FR milk for OA/C18:0 ratio was not statistically significant (Table 1). There were no significant differences in FA profile between CON and FR milk (Table 1). The effect of production system in the full FA profile of milk (80 individual FA) is shown in the Appendix (Table A1).

### *3.2.2 Effect of month*

Significant effects of month were identified for milk concentrations of all individual FA (except DHA) and FA groups and indices (Table 2). Concentrations of C12:0 in milk fat were lower in June-October than in March-May and November-February, with numerically smaller significant differences between months within these periods also being observed (Table 2). Concentrations of C14:0 and SFA and the AI and TI in milk were lower in May-October than in March and November-February, with numerically smaller significant differences between months within these periods also being observed; their values were intermediate in April and lower when compared with March and November-February (Table 2). Milk contained more C16:0 in March and October-February than in May-August and had intermediate concentrations in April and September, which were also lower when compared with October-February (Table 2). Milk contained more C18:0 in May-September than in March and November-February, and had intermediate concentrations in April and October (Table 2). Concentrations of VA, ALNA and EPA and n-3/n-6 in milk were higher in May-October than in March-April and November-February, with numerically smaller significant differences between months within these periods also being observed (Table 2). OA concentrations and  $\Delta^9$ I in milk were higher in May-October than in March and November-February, with numerically smaller significant differences between months within these periods also being observed; their values were intermediate in April (Table 2). Milk contained more LA in March-May and October, than in July and November-February, with the remaining months showing intermediate values, mostly without being significantly different (Table 2). RA and *trans* FA

188 concentrations in milk were highest in May-September, lowest in March and November-  
189 February, and showed intermediate values in April and October, with numerically smaller  
190 significant differences between months within these periods also being observed (Table 2).  
191 DPA concentrations in milk were higher in May-November than in March-April and  
192 December-February, with numerically smaller significant differences between months within  
193 these periods also being observed; the means for these parameters were intermediate in  
194 January-February and higher when compared with March-April (Table 2).

195 Concentrations of MUFA, *cis* MUFA, *trans* MUFA, PUFA, *cis* PUFA, *trans* PUFA, *cis/trans*  
196 plus *trans/cis* PUFA and n-3 in milk were higher in May-October than in March and  
197 November-February, with numerically smaller significant differences between months within  
198 these periods also being observed; the means for these parameters were intermediate in April  
199 (Table 2). Milk contained more n-6 in March-May and September-October, than in July and  
200 November-January, with the remaining months showing intermediate values, mostly without  
201 being significantly different (Table 2). When excluding VA, *trans* FA concentrations in milk  
202 were highest in May-July, lowest in September-November, and showed intermediate values in  
203 March-April, August and December-February; numerically smaller significant differences  
204 between months within these periods were also observed (Table 2).

205 Ratio of C14:1/C14:0 was higher in September-November than in March-June and February,  
206 with numerically smaller significant differences between months within these periods also  
207 being observed; the means for these parameters were intermediate in July-August and  
208 December-January and higher when compared with March-May (Table 2). Ratio of  
209 C16:1/C16:0 in milk was highest in May-October, lowest in March and December-February,  
210 and showed intermediate values in April and November, with numerically smaller significant  
211 differences between months within these periods also being observed (Table 2). Ratio of  
212 OA/C18:0 in milk was highest in September-November, lowest in April and December-

February, and showed intermediate values in March and May-August, although the difference between March and April and December-February was not statistically significant; numerically smaller significant differences between months within these periods were also observed (Table 2). Milk had higher ratio of RA/VA in June-August and October-December than in May, with the remaining months showing intermediate values, mostly without being significantly different (Table 2). The effect of month in the full FA profile of milk (80 individual FA) is shown in the Appendix (Table A2).

### 3.2.3 Effect of the production system $\times$ month interaction

Significant effects of the production system  $\times$  month interaction were identified for milk concentrations of C12:0, C16:0, LA, RA, ALNA (Figure 1), SFA, MUFA, PUFA, n-6, *trans* FA (total or by excluding VA), n-3/n-6 ratio and AI (Figure 2),  $\Delta^9$ I, C14:1/C14:0, C16:1/C16:0, OA/C18:0 and VA/RA (Appendix, Figure A2). C12:0 concentrations in ORG milk were lower than in CON and FR milk in March, but higher than in FR milk in May; C12:0 concentrations in FR milk were higher than in ORG and CON milk in July and December-February (Figure 1a). C16:0 concentrations were lower in ORG milk than in CON milk in April-November, and compared with FR milk in February; concentrations in FR milk were also lower than in CON milk in May and August-September (Figure 1b).

LA concentrations were higher in ORG milk than in FR milk in April-May and higher in CON milk than in FR milk in December (Figure 1c). ORG milk had higher RA concentrations when compared with FR and CON milk throughout the year, although differences were not statistically significant between ORG and FR milk in August-September and between ORG and CON milk in January-February; FR milk had higher RA concentrations than CON milk in September (Figure 1d). ORG milk had higher ALNA concentrations when compared with FR and CON milk throughout the year; FR milk had higher ALNA concentrations than CON milk in May and August-September (Figure 1e).

SFA concentrations were lower in ORG milk than in FR milk in April, October and February, and when compared with CON milk in April-May; when compared with CON milk, FR milk had less SFA in May but more SFA in February (Figure 2a). FR milk had lower MUFA concentrations than ORG milk in May and CON milk in February, and higher MUFA concentrations than CON milk in June (Figure 2b). ORG milk had higher *trans* MUFA concentrations than FR and CON milk in April-November but differences in April-May and November were not significant when compared with FR and CON milk, respectively (Figure 2c). ORG milk had higher PUFA concentrations when compared with FR and CON milk throughout the year; CON milk had higher PUFA concentrations than FR milk in November-February (Figure 2d). ORG milk had higher concentrations of *cis* PUFA than FR milk in April-July and October-February, and when compared with CON milk in April-May, July, November-December and February; CON milk also contained more *cis* PUFA than FR milk in November-December and February (Figure 2e). FR milk had lower n-6 concentrations compared with ORG milk in May and with CON milk in November-December and February; CON milk had higher LA concentrations than ORG milk in September (Figure 2f). ORG milk had higher concentrations of *trans* FA in April-July and October, than CON and FR milk (Figure 2g). When excluding VA from *trans* FA, ORG milk had higher concentrations than FR milk in April, and when compared with CON milk in July; FR milk contained less *trans* FA (excluding VA) than CON milk in April, November and December (Figure 2h). ORG milk had higher n-3/n-6 when compared with FR and CON milk in June-February, and when compared with CON milk in April-May; FR milk also had higher n-3/n-6 than CON milk in May (Figure 2i). AI was higher in FR milk than in ORG milk in April and February and higher than in CON milk in February; ORG milk had lower AI than CON milk in April (Figure 2j).

### 3.3 Estimated fatty acid intakes

When the effect of production system on the estimated FA intakes (according to the National Diet and Nutrition survey (Bates, et al., 2014) and milk FA profiles measured in the present study) was assessed, significant effects were identified for the intakes of PUFA, n-3, ALNA, EPA+DHA, and *trans* FA across all age groups and genders (Table 3). In male children 4-10 years old (yo), estimated intakes from ORG milk fat were higher for PUFA (+67 and +85 mg/d), n-3 (+51 and +41 mg/d), ALNA (+31 and +26 mg/d), EPA+DHA (+3 and +2 mg/d), and *trans* FA (+57 and +70 mg/d), when compared with CON and FR milk fat, respectively. In male teenagers 11-18 yo, estimated intakes from ORG milk fat were higher for PUFA (+60 and +77 mg/d), n-3 (+46 and +47 mg/d), ALNA (+28 and +24 mg/d), EPA+DHA (+2 and +2 mg/d), and *trans* FA (+52 and +63 mg/d), when compared with CON and FR milk fat, respectively. In adult males 19-64 yo, estimated intakes from ORG milk were higher for PUFA (+61 and +77 mg/d), n-3 (+46 and +37 mg/d), ALNA (+28 and +24 mg/d), EPA+DHA (+2 and +2 mg/d), and *trans* FA (+52 and +64 mg/d), when compared with CON and FR milk fat, respectively. In adult males over 65 yo, estimated intakes from ORG milk were higher for PUFA (+77 and +99 mg/d), n-3 (+58 and +47 mg/d), ALNA (+36 and +30 mg/d), EPA+DHA (+3 and +3 mg/d), and *trans* FA (+66 and +81 mg/d), when compared with CON and FR milk fat, respectively. In female children 4-10 yo, estimated intakes from ORG milk were higher for PUFA (+63 and +81 mg/d), n-3 (+48 and +39 mg/d), ALNA (+30 and +25 mg/d), EPA+DHA (+3 and +2 mg/d), and *trans* FA (+54 and +66 mg/d), when compared with CON and FR milk fat, respectively. In female teenagers 11-18 yo, estimated intakes from ORG milk fat were higher for PUFA (+49 and +62 mg/d), n-3 (+37 and +30 mg/d), ALNA (+23 and +19 mg/d), EPA+DHA (+2 and +1 mg/d), and *trans* FA (+42 and +51 mg/d), when compared with CON and FR milk fat, respectively. In adult females 19-64 yo, estimated intakes from ORG milk were higher for PUFA (+42 and +54 mg/d), n-3 (+32 and +26 mg/d), ALNA (+20 and +17 mg/d), EPA+DHA (+2 and +1 mg/d), and *trans* FA (+36 and +44 mg/d), when compared with

CON and FR milk, respectively. In adult females over 65 yo, estimated intakes from ORG milk were higher for PUFA (+66 and +84 mg/d), n-3 (+50 and +40 mg/d), ALNA (+30 and +25 mg/d), EPA+DHA (+3 and +2 mg/d), and *trans* FA (+56 and +68 mg/d), when compared with CON and FR milk fat, respectively.

## 4. Discussion

### 4.1 Milk basic composition

ORG milk contained more fat than CON milk (as in previous UK retail studies; (Butler, et al., 2011b)), and FR milk. This may be an effect of fat standardisation at processing plants rather than an effect of production system, as this is common practice in CON and FR, but not in ORG, supply chain. Although the effect of production system on lactose content was significant, the numerical differences were marginal (0.4 g/kg less in ORG than in CON and FR milk) and potential relevance to consumer health is small. This difference may be due to fat standardisation in CON and FR milk; removing fat from whole milk decreases the dilution factor for remaining milk solids, so could increase milk lactose concentration when expressed as g/kg of milk. A significant effect of production system was not observed, for milk protein, casein and SCC contents, in agreement with previous reports for UK milk (Butler, et al., 2011b; Stergiadis, et al., 2012).

### 4.2 Milk fatty acid profile

#### 4.2.1 Organic milk

The finding that ORG milk contains similar concentrations of SFA to CON milk is consistent with previous UK retail (Butler, et al., 2011b) and farm (Ellis, et al., 2006; Stergiadis, et al., 2012) surveys. In the current study, ORG milk had lower SFA concentrations during the period when cows are turned out to graze (spring) in UK, but also in October and December. However,



the principal SFA in milk fat, C16:0, which is considered undesirable in human nutrition, was found in lowest concentrations in ORG milk, in line with a previous UK retail study (Butler, et al., 2011b). Milk C16:0 originates both from diet and endogenous synthesis by the mammary gland (Chilliard, et al., 2007). Concentration of C16:0 in ORG milk may reflect that lipids in ORG cow diets (rich in fresh grass and with a high forage:concentrate ratio) may contain proportionately less C16:0 than conventional cow diets; and/or cause a potential modification in the amounts of the C16:0 substrates in the rumen, which are used for its *de novo* synthesis in the mammary gland (Chilliard, et al., 2007). Multivariate analyses in other studies (Stergiadis, et al., 2015a; Stergiadis, et al., 2015b; Stergiadis, et al., 2012) have shown a negative relationship between fresh forage intake (which is expected to be higher in ORG systems), and milk C16:0 concentration. Conversely, other studies reported a higher 16:0 concentration (USA; (O'Donnell, Spatny, Vicini, & Bauman, 2010)) or no difference (the Netherlands; (Capuano, et al., 2015)) between organic and conventional milk. As cow diet is a major driver of milk FA profile (Stergiadis, et al., 2015a; Stergiadis, et al., 2015b; Stergiadis, et al., 2012), these discrepancies probably arise due to variations in diets used in different countries as result of contrasting soil, climate, tradition and legislation (Butler, et al., 2011a). Milk MUFA concentrations, mainly characterised by the nutritionally-desirable OA which represented 73.1% of total MUFA in the present work, were similar between ORG and CON milk, thus agreeing with previous UK studies (Butler, et al., 2011b; Stergiadis, et al., 2012). The concentrations of OA may be affected by dietary supply, extent of rumen biohydrogenation (RBH), but also its synthesis from C18:0 by  $\Delta^9$ -desaturase action in the mammary gland (Destailats, Trottier, Galvez, & Angers, 2005). Differences between ORG and CON systems in these factors may have been either small or showing a counteracting effect, thus resulting in similar concentrations of OA in ORG and CON milk.

334 The finding that ORG milk contains more VA and RA than CON milk is in line with previous  
335 retail surveys (Butler, et al., 2011b; Capuano, et al., 2015; O'Donnell, et al., 2010), although  
336 other UK farm surveys have shown either no differences (Ellis, et al., 2006) or a significant  
337 difference only when ORG was compared with intensive CON production systems during  
338 summer (Stergiadis, et al., 2012). VA is an intermediate product of RBH of dietary PUFA, and  
339 in particular ALNA (Chilliard, et al., 2007; Destailats, et al., 2005). Upon absorption and  
340 delivery to the mammary gland, part of VA is converted to RA, under the effect of mammary  
341  $\Delta^9$ -desaturase (Chilliard, et al., 2007; Destailats, et al., 2005). Therefore, cow diets rich in  
342 ALNA, such as those of high pasture intake characterizing ORG systems, will increase the  
343 availability of substrate for higher VA production in the rumen and the subsequent RA  
344 synthesis in the mammary gland (Chilliard, et al., 2007; Elgersma, 2015). Pasture intake is  
345 potentially the main driver for milk VA and RA concentrations in the current study because  
346 differences in RA concentrations between ORG and CON were not significant during the  
347 period that pasture was not available in the UK (December to February).

348 The higher concentrations of ALNA, EPA and DPA, and consequently the n-3, in ORG than  
349 in CON milk is in line with other retail surveys (Butler, et al., 2011b; Capuano, et al., 2015;  
350 O'Donnell, et al., 2010). However, for DPA, retail surveys may show only a tendency for higher  
351 concentrations in ORG (Butler, et al., 2011b) or inconsistent results between summer and  
352 winter (Capuano, et al., 2015). In a previous UK farm survey, ORG milk contained more DPA  
353 only when compared with milk from highly-intensive CON production systems (Stergiadis, et  
354 al., 2012). The higher concentrations of ALNA in ORG milk were observed throughout the  
355 year. In summer, milk ALNA concentrations can be enhanced by (i) higher pasture intake in  
356 ORG systems, due to the higher ALNA supply from fresh forage than conserved forage and/or  
357 concentrates (Elgersma, 2015), and (ii) clover contribution to the grazing swards, potentially  
358 due to the increased transfer rates of dietary ALNA when fresh clover substitutes fresh grass

359 in cow diets (Stergiadis, et al., 2018). In winter, grass/clover silage commonly used in ORG  
360 systems when pasture is not available, has been found to increase ALNA concentrations when  
361 compared with grass or grass/maize silage (Dewhurst, Fisher, Tweed, & Wilkins, 2003;  
362 Wiking, Theil, Nielsen, & Sorensen, 2010), commonly used in the CON systems. This may be  
363 explained by the slower DM degradation (Dewhurst, Evans, Scollan, Moorby, Merry, &  
364 Wilkins, 2003) and rates of RBH of clover compared with grass (Lejonklev, Storm, Larsen,  
365 Mortensen, & Weisbjerg, 2013), which may increase rumen passage rates, reduce RBH of  
366 ALNA, and eventually increase transfer rates of dietary ALNA to milk. In humans and animals,  
367 including cattle, ALNA is used as substrate for the synthesis of EPA and DPA by various  
368 enzymes, including elongases,  $\Delta^5$ -desaturase and  $\Delta^6$ -desaturase (Barcelo-Goblijn, et al., 2009).  
369 Therefore, a higher supply of ALNA in the mammary gland of cows in the ORG systems, as a  
370 consequence of the combined effect of high pasture and clover intake, may have increased the  
371 substrate available for EPA and DPA synthesis.

372 Factors affecting  $\Delta^9$ -desaturase activity, potentially including, animal genetics, production  
373 stage and diet, are not well understood. Transition from winter diets (relying on conserved  
374 forages and concentrates) to summer diets (including substantial amounts of pasture) is likely  
375 to (i) alter the supply of FA, transferred directly to milk or acting as substrate for conversion  
376 to other milk FA, and (ii) exert metabolic changes in the rumen and/or the cow, possibly  
377 altering the activity of nutritionally-sensitive enzymes responsible for *de novo* synthesis of  
378 short and medium chain SFA or desaturation of FA in the mammary gland (Lock &  
379 Garnsworthy, 2003). Higher fresh grass intakes, which increase water-soluble carbohydrate  
380 intakes and the subsequent insulin levels, may increase  $\Delta^9$ -desaturase activity (Lock, et al.,  
381 2003), but the opposite was observed in the ORG milk in the current study. ORG dairy herds  
382 in the UK extensively use crossbred cows (most typically crosses between Holstein,  
383 British/New Zealand Friesian, Jersey and/or Scandinavian Red) while CON herds rely almost

entirely in Holstein cows. Provided the well documented substantial effect of breed and individual differences on  $\Delta^9$ -desaturase activity, such as the lower activity in Jersey and Holstein  $\times$  Jersey crosses than in pure Holstein cows (Palladino, Buckley, Prendiville, Murphy, Callan, & Kenny, 2010), it is possible that the effect of breed may have overridden any potential effect of diet.

#### *4.2.2 Potential impact of organic milk on the fatty acid intakes of UK consumers*

Department of Health and Social Care in the UK has set a maximum reference nutrient intakes (RNI) of SFA and *trans* FA at 11% and 2% of food energy intake, respectively, and recommended intakes of PUFA at 6.5% of food energy intake (Department of Health, 1991). According to the current average requirements for energy for the different age groups and genders, and an energy content of fat at 37 kJ/g (SACN, 2011), under the current dairy fat intakes in the UK (Bates, et al., 2014), milk fat contributes 24-37% of the maximum recommended intakes of SFA intakes in adults (being maximum for men over 65 yo, which is the group with the maximum milk fat intakes) and 20-43% of the maximum recommended intakes in children. Overall intakes of SFA will not be affected by switching between milks from different production systems because the effect of production system on milk SFA concentrations was not significant. Even within months that the difference between ORG and CON was maximised and was statistically significant (April; milk SFA concentrations of 680.4 and 705.2 g/kg FA respectively), consumption of ORG milk would minimally reduce SFA intake by 0.250 g/d in men over 65 yo (contributing 36.5% RNI than 37.4% RNI) and 0.214 g/d in women (contributing 36.7% RNI than 37.6% RNI), when compared with CON milk. Previous work has suggested that in order to benefit public health and reduce health care costs, a reduction on milk SFA should be at the level of 150 g/kg FA (Kliem, et al., 2013). According to these, any potential public health impact from the occasional reduction in milk SFA intakes via consumption of ORG milk cannot be claimed by the results of the present work.

Previous work has highlighted that some milk SFA included in the calculations of total milk SFA (e.g. C4:0, C8:0, C10:0), may have beneficial implications to human health (Haug, et al., 2007). Therefore, focusing on SFA which, if excessively consumed, increase CVD risk (C12:0, C14:0 and C16:0; (FAO, 2010)), may be more accurate when discussing milk fat profile. According to the results of this survey and current dairy food consumption in the UK (Bates, et al., 2014), switching from CON to ORG milk fat will reduce consumption of C16:0 by 0.206 g/d (from 4.0 to 3.8 g/d) in children 4-10 yo, by 0.164 g/d (from 3.2 to 3.0 g/d) in children 11-18 yo, by 181 mg/d (from 3.5 to 3.3 g/d) in adults 19-64 yo, and by 243 mg/d (from 4.7 to 4.5 g/d) in adults over 65 yo. Nutritional recommendations are provided only for total SFA (Department of Health, 1991), so it is not possible to estimate C16:0 contribution from dairy foods.

According to the results of the present survey and the current dairy fat intakes in the UK (Bates, et al., 2014), switching from CON to ORG milk fat will increase the contribution of PUFA, expressed as % RNI, from 4.0% to 4.6% in children 4-10 yo, from 2.1% to 2.7% in children 11-18 yo, from 2.4% to 2.9% of adults 19-64 yo, and from 3.6% to 4.4% in adults over 65 yo. The maximum benefit to PUFA intakes from switching from CON to ORG milk was observed in May (milk PUFA concentrations 52.1 g/kg FA for ORG and 43.7 g/kg FA for CON) when intakes were increased by 131 mg/d in men over 65 yo (contributing 4.0% RNI than 4.7% RNI), and 111 mg/d in women over 65 yo (contributing 4.0% RNI than 4.8% RNI). Although the potential health effects by these changes have not been investigated in the current study, these differences are rather small and are unlikely to be associated with reduced chronic disease risk within a whole diet.

A switch to ORG milk would increase the intakes of *trans* FA, expressed as % RNI, from 10.4% to 11.8% in children 4-10 yo, from 6.0% to 6.8% in children 11-18 yo, from 6.7% to 7.6% of adults 19-64 yo, and from 10.1% to 11.4% in adults over 65 yo. However, in this study

434 43% of *trans* FA in milk fat was VA, which is associated with positive effects in human health  
435 (Field, et al., 2009). If concentrations of VA are not included in the calculation of *trans* FA,  
436 the overall intakes will not be affected by switching between milks from different production  
437 systems because the effect of production system on milk *trans* FA (excluding VA)  
438 concentrations was not statistically significant.

439 The European Food and Safety Authority set the adequate intake (ADI) for ALNA at 0.5%  
440 energy intake (EFSA, 2010). According to the results of the present survey and the current  
441 dairy fat intakes in the UK (Bates, et al., 2014), switching from CON to ORG milk fat will  
442 increase the intakes of ALNA from 5.8% to 8.9% ADI in children 4-10 yo, from 3.0% to 4.6%  
443 ADI in children 11-18 yo, from 3.4% to 5.2% ADI in adults 19-64 yo and from 5.2% to 7.9%  
444 ADI in adults over 65 yo. The maximum potential benefit in intakes of ALNA was observed  
445 in September (milk ALNA concentrations 7.8 g/kg FA for ORG and 4.7 g/kg FA for CON)  
446 when intakes could have been increased by 48 mg/d in males over 65 yo (contributing 9.2%  
447 RNI than 5.6% ADI), and 40.8 mg/d in females over 65 yo (contributing 9.3% ADI than 5.6%  
448 ADI), when compared with CON milk.

449 The European Food and Safety Authority set the adequate intake (ADI) for EPA+DHA of 250  
450 mg/d in adults and children over 24 months old (EFSA, 2010). According to the results of this  
451 survey and the current milk intakes in the UK (Bates, et al., 2014), switching from CON to  
452 ORG milk fat will increase the intakes of EPA+DHA from 2.7% ADI to 3.6% ADI in children  
453 4-10 yo, from 2.1% ADI to 2.9% ADI in children 11-18 yo, from 2.7% to 3.2% ADI in adults  
454 19-64 yo, and from 3.2% to 4.3% ADI in adults over 65 yo. The maximum potential benefit in  
455 intakes of EPA+DHA was observed in June (milk EPA+DHA concentrations 0.84 g/kg FA for  
456 ORG and 0.78 g/kg FA for CON) when intakes could have been increased by 4.0 mg/d in males  
457 over 65 yo (contributing 5.2% ADI than 3.6% ADI), and 3.4 mg/d in females over 65 yo  
458 (contributing 4.4% ADI than 3.1% ADI), when compared with CON milk. Part of dietary

ALNA in humans is converted to EPA, DPA and DHA, but with conversion efficiencies being lower than 0.2% (Barcelo-Goblijn, et al., 2009). An additional supply of EPA+DHA may therefore be expected, as a result of milk ALNA metabolism, but the low conversion efficiencies, will result in minimal additional supply of EPA+DHA from ORG than CON milk (less than 1 mg/d).

According to the results of this survey and the current milk intakes in the UK (Bates, et al., 2014), switching from CON to ORG milk fat will increase RA intake by 24.7 mg/d (from 71.5 to 96.2 mg/d) in children 4-10 yo, by 19.6 mg/d (from 56.9 to 76.5 mg/d) in children 11-18 yo, by 21.6 mg/d (from 62.6 to 84.2 mg/d) in adults 19-64 yo, and by 29.1 mg/d (from 84.2 to 113.3 g/d) in adults over 65 yo. Given that on average 19 % of VA is also endogenously converted to RA in the human body (Field, et al., 2009), consumption of ORG milk may also increase the available RA via endogenous synthesis, because of its higher VA concentrations. RA has been previously associated with a number of health benefits in humans (Dilzer, et al., 2012). However, nutritional recommendations for RA are not currently developed, so it is not possible to estimate RA contribution from dairy foods.

Overall, organic milk could be considered desirable from a human nutrition perspective, in terms of lower SFA and higher PUFA, n-3, ALNA, RA contents, as well as a higher ratio of n-3/n-6 and lower TI (which however refer to the whole diet), thus aligning with current nutritional recommendations (EFSA, 2010; FAO, 2010; Givens, 2017). A switch from CON to ORG milk will influence the intakes of these FA, but any implications for human health cannot be drawn in the present study, because these changed intakes are relatively small. The nutritional recommendations (Department of Health, 1991; EFSA, 2010; SACN, 2011) for individual FA or FA groups refer to the total diet rather than a single food, and although the current study estimates the potential changes on FA intakes from dairy products, any potential effect on human health will be influenced by FA intakes from other foods. Current evidence

suggests there is no positive association between intake of milk and dairy products and the risk of CVD and type-2 diabetes, while consumption of cheese and yoghurt also showed a negative association (Kliem, et al., 2011; Thorning, et al., 2017). This may be an effect of interactions between milk matrix components and enhances the necessity for future research on the effect of milk and dairy products as whole foods, alongside investigations of the nutritional role of their individual components (Kliem, et al., 2011; Thorning, et al., 2017).

#### *4.2.3 Free-range milk*

The lack of differences between the two conventional systems (CON, FR), one representing typical CON UK dairy management and the other FR practices, is in line with recent results from the Netherlands (Capuano, et al., 2015). Provided that diet, and in particular fresh grass intake, is the major driver for milk FA profiles (Elgersma, 2015; Stergiadis, et al., 2015b; Stergiadis, et al., 2012), the similarities between CON and FR milk may potentially reflect small differences in cow nutrition between the two production systems. Previous studies that showed substantial differences in the FA profile of milk between pasture-based, ORG and CON milk at farm level had investigated low-input pasture-based farms where the average pasture intake was more than 95% of total cow diet (Butler, et al., 2008; Stergiadis, et al., 2015b). Provided that FR certification refers to access to outdoors/pasture but without setting minimum requirements for pasture intake, lower contribution of pasture in cow diet when compared with the low-input farms assessed in other studies (Butler, et al., 2008; Stergiadis, et al., 2015b), may potentially explain the lack of effect on milk FA profile. In addition, allowing cows access to pasture for six months is a typical practice in UK CON dairy systems, although maybe at a lesser extent than in FR systems, and this may further contribute to the similarities in cow diets and the subsequent FA profile between CON and FR milk. Other potential reasons for this observations that have been provided in the Netherlands was (i) that not all farms provide access to pasture the same time, thus diluting the effect of pasture intake when bulking milk at



the dairy, and (ii) the contribution of fresh-cut grass in indoor CON systems, which would reduce the differences between grazing and indoor cows (Capuano, et al., 2015). Despite the overall similarities between FR and CON milk, significant differences were observed in specific months within the grazing season, potentially as an effect of the higher pasture intake in FR herds than in CON herds during these months. For example, FR milk had a more preferable FA profile than CON milk in May, August and/or September, by containing less of the nutritionally-undesirable C16:0, more of the nutritionally-desirable ALNA and RA and by having a higher n-3/n-6 ratio; thus representing a favourable effect of substituting SFA with MUFA and PUFA, in line with current recommendations (EFSA, 2010; FAO, 2010; Givens, 2017). FR milk also contained less SFA and more MUFA in May, but this relationship was reversed in February. The lower PUFA in FR than CON milk during the indoor period mainly reflects the lower concentrations of n-6 and LA, which may be a result of lower use of maize silage, a main driver for milk n-6 concentrations (Chilliard, et al., 2007; Stergiadis, et al., 2015b), in FR than in CON herds. Inconsistency in the differences between FR and CON milk throughout the year may have also been a consequence of the small number of farms contributing the FR milk at the dairies. Because of that, the effect of cow diet in individual FR farms has a proportionately higher impact to the final product.

#### *4.2.4 Seasonal variation*

The effect of season on the FA profile of UK retail milk has been extensively investigated in other surveys (Butler, et al., 2011b; Kliem, et al., 2013). In agreement with the present work, these previous studies highlighted that concentrations of milk total SFA and individual SFA (C12:0, C14:0, C16:0) are lower, and those of VA, OA, RA, ALNA, EPA, n-3, PUFA, EPA are higher, during the grazing season. In the UK, in dairy systems where cows have access to pasture, and in line with local climate, animals are housed in winter (December-February), turn out to pasture at March-April, have a period where grazing is maximised (May-September) and

then gradually return to indoor diets rich in conserved forages and concentrates late in autumn (October-November). Fresh grass is rich in PUFA, including ALNA, and therefore its potentially higher dietary intakes may result in (i) higher milk ALNA concentrations, via direct transfer to milk (Elgersma, 2015), (ii) higher OA, via rumen synthesis of C18:0 (end product of RBH of FA) and the subsequent increased C18:0 supply in the mammary gland for OA synthesis (Destailats, et al., 2005), (iii) higher milk VA and RA concentrations, via rumen synthesis of VA and the subsequent increase in VA supply in the mammary gland for RA synthesis (Chilliard, et al., 2007; Destailats, et al., 2005), (iv) higher EPA and DPA, potentially via the higher supply of their substrate ALNA (Barcelo-Goblijn, et al., 2009), and eventually (v) lower total SFA and individual SFA (C12:0, C14:0, C16:0) concentrations. Given that 60% of the samples in the present study were from ORG and FR farms, which are expected to provide access to pasture during the grazing season, the effect of pasture intake appears to be the most possible explanation of the seasonal variation. The fact that pasture intake is among the strongest drivers of milk FA profiles in the UK, has been previously demonstrated in multivariate redundancy analyses in data collected from dairy farms (Stergiadis, et al., 2015b; Stergiadis, et al., 2012).

## 5. Conclusions

Organic retail milk showed a more favourable FA profile, containing more nutritionally-desirable FA, less C16:0, and a higher n-3/n-6 ratio than conventional milk. During specific months, organic milk also had less total SFA. The free-range milk had similar FA profile to conventional milk, but contained less SFA (including C16:0) and more ALNA and RA, in specific months within the outdoor/grazing season. Although background information on dairy management practices was not available, it is highly likely that differences in milk FA profiles resulted from contrasting cow diets, and in particular the intakes of pasture, clover and forage:concentrate ratio. Based on measured milk FA profiles across the production systems,

559 dairy fat contributes approximately one-third of the maximum recommended intake of SFA in  
560 adult consumer diets. Consuming organic dairy products would increase intakes of  
561 nutritionally-desirable PUFA, and reduce consumption of nutritionally-undesirable SFA.  
562 However, when compared with conventional milk in terms of daily recommended intakes of  
563 these FA, there would be relatively little difference. Therefore, any implications to human  
564 health cannot be drawn from the present study.

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**Table 1**

Means (and average SE) and ANOVA P-values for the effect of production system (conventional, CON; organic, ORG; free-range, FR) on the basic composition and fatty acid (FA) profile (g/kg total FA) of milk collected from retail outlets during the year

Parameters assessed	Production System			SE	ANOVA P-values <sup>b</sup>
	CON n=48	ORG n=48	FR n=24 <sup>a</sup>		
<b>Basic composition</b>					
Fat (g/kg milk)	34.9 <sup>B</sup>	40.0 <sup>A</sup>	37.0 <sup>B</sup>	0.60	***
Protein (g/kg milk)	32.7	32.4	32.7	0.49	ns
Casein (g/kg milk)	25.5	25.2	25.4	0.41	ns
Lactose (g/kg milk)	45.2 <sup>A</sup>	44.8 <sup>B</sup>	45.2 <sup>A</sup>	0.10	*
SCC <sup>c</sup> (x 10 <sup>3</sup> /ml milk)	38	137	58	30	ns
<b>Individual FA</b>					
<b>SFA</b> <sup>d</sup>					
C12:0	33.4	32.4	34.8	0.67	ns
C14:0	111	114	114	1.2	ns
C16:0	331 <sup>A</sup>	314 <sup>B</sup>	325 <sup>A</sup>	2.0	***
C18:0	99.5	106.1	103.3	2.36	ns
<b>MUFA</b> <sup>e</sup>					
VA	12.2 <sup>B</sup>	17.1 <sup>A</sup>	12.3 <sup>B</sup>	0.68	**
OA	200	197	199	2.6	ns
<b>PUFA</b> <sup>f</sup>					
LA	17.1	16.6	15.3	0.75	ns
RA	5.91 <sup>B</sup>	7.95 <sup>A</sup>	6.06 <sup>B</sup>	0.259	***
ALNA	4.39 <sup>B</sup>	6.71 <sup>A</sup>	4.76 <sup>B</sup>	0.124	***
EPA	0.484 <sup>B</sup>	0.674 <sup>A</sup>	0.551 <sup>B</sup>	0.0226	***
DPA	0.795 <sup>B</sup>	1.024 <sup>A</sup>	0.834 <sup>B</sup>	0.0279	***
DHA	0.067	0.078	0.060	0.0100	ns
<b>FA groups</b>					
SFA	688	684	692	2.6	ns
MUFA	273	272	270	2.4	ns
<i>cis</i> MUFA <sup>g</sup>	242	237	240	2.9	ns
<i>trans</i> MUFA <sup>h</sup>	31.0 <sup>B</sup>	35.0 <sup>A</sup>	30.0 <sup>B</sup>	0.84	***
PUFA	39.7 <sup>B</sup>	44.7 <sup>A</sup>	38.3 <sup>B</sup>	0.42	***
<i>cis</i> PUFA <sup>i</sup>	25.9 <sup>B</sup>	28.0 <sup>A</sup>	24.4 <sup>B</sup>	0.31	*
<i>trans</i> PUFA <sup>j</sup>	0.34 <sup>B</sup>	0.52 <sup>A</sup>	0.37 <sup>B</sup>	0.033	*
<i>cis/trans</i> + <i>trans/cis</i> PUFA <sup>k</sup>	13.4 <sup>B</sup>	16.1 <sup>A</sup>	13.5 <sup>B</sup>	0.49	**
n-3 <sup>l</sup>	7.93 <sup>B</sup>	11.69 <sup>A</sup>	8.66 <sup>B</sup>	0.332	***
n-6 <sup>m</sup>	20.9	20.0	18.9	0.81	ns
n-3/n-6	0.39 <sup>B</sup>	0.59 <sup>A</sup>	0.46 <sup>B</sup>	0.031	**
<i>trans</i> FA <sup>n</sup>	3.13 <sup>B</sup>	3.55 <sup>A</sup>	3.03 <sup>B</sup>	0.086	**
<i>trans</i> FA (exc. VA)	1.90	1.84	1.81	0.043	ns
<b>Indices</b>					
<i>Human health-related</i>					
AI <sup>o</sup>	2.60	2.56	2.69	0.047	ns
TI <sup>p</sup>	3.13 <sup>A</sup>	2.89 <sup>B</sup>	3.15 <sup>A</sup>	0.027	***
<i><math>\Delta^9</math>-desaturase activity</i>					
$\Delta^9$ I <sup>r</sup>	0.297	0.296	0.296	0.0026	ns
C14:1/C14:0	0.084 <sup>A</sup>	0.080 <sup>B</sup>	0.084 <sup>A</sup>	0.0007	**
C16:1/C16:0	0.058 <sup>A</sup>	0.056 <sup>B</sup>	0.058 <sup>A</sup>	0.0006	*
OA/C18:0	2.011 <sup>A</sup>	1.856 <sup>B</sup>	1.928 <sup>A</sup>	0.0371	*
RA/VA	0.491	0.469	0.501	0.0085	†

<sup>a</sup> In September, there was a missing sample of free-range milk in the analysis of basic composition and the mean on this set of parameters was calculated from 23 samples

<sup>b</sup> Significances were declared at \*\*\*, P < 0.001; \*\*, P < 0.01; \*, P < 0.05; †, 0.05 < P < 0.10 (trend); ns, P > 0.10 (non-significant). Means for production system within a row with different upper case letters are significantly different according to Fisher's Least Significant

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Difference test ( $P < 0.05$ )

<sup>c</sup> Somatic cell count

<sup>d</sup> Saturated FA: C4:0, C5:0, C6:0, C7:0, C8:0, C9:0, C10:0, C11:0, C12:0, C13:0, C13:0*iso*, C13:0*anteiso*, C13:0, C14:0*iso*, C14:0, C15:0*anteiso*, C15:0, C16:0*iso*, C16:0, C17:0*iso*, C17:0, C18:0*iso*, C18:0, C20:0, C22:0, C24:0

<sup>e</sup> Monounsaturated FA (MUFA): *c9* C10:1, *c10* C11:1, *c9* C12:1, *c9* C13:1, *t9* 14:1, *c9* C14:1, *c10* C15:1, *t7+t8* C16:1, *t9* C16:1, *t11+t12+t13* C16:1, *c9* C16:1 (co-elutes with C17:0*anteiso*), *c11* C16:1, *c13* C16:1, *t10* C17:1, *c9* C17:1, *t4* C18:1, *t5* C18:1, *t6+t7+t8* C18:1, *t9* C18:1, *t10* C18:1, *t11* C18:1 (VA), *c6+t12* C18:1, *c9* C18:1 (OA), *t15* C18:1, *c11* C18:1, *c12* C18:1, *c13* C18:1, *t16 + c14* C18:1, *c15* C18:1 (co-elutes with C19:0), *c16* C18:1, *c5* C20:1, *c8* C20:1, *c11* C20:1, *c13* C22:1, *c15* C24:1

<sup>f</sup> Polyunsaturated FA (PUFA): *t11t15* C18:2, *t9t12* C18:2, *c9t13* C18:2, *c10t14* C18:2, *c9t14* C18:2, *c9t12* C18:2, *t9c12* C18:2, *t11c15* C18:2, *c9c12* C18:2 (LA), *t12c15* C18:2 (co-elutes with *c9* C19:1), *c6c9c12* C18:3, *c9c12c15* C18:3 (ALNA), *c9c11* C18:2 conjugated (RA) (co-elutes with *t7c9+t8c10+t6c8* C18:2), other C18:2 conjugated FA of unknown isomerism, *c11c14* C20:2, *c8c11c14* C20:3, *c11c14c17* C20:3, *c5c8c11c14* C20:4, *c13c16* C22:2, EPA, *c13c16c19* C22:3, DPA, DHA

<sup>g</sup> *cis* MUFA: *c9* C10:1, *c10* C11:1, *c9* C12:1, *c9* C13:1, *c9* C14:1, *c10* C15:1, *c9* C16:1 (co-elutes with C17:0*anteiso*), *c11* C16:1, *c13* C16:1, *c9* C17:1, *c6* C18:1 (co-elutes with *t12* C18:1), OA, *c11* C18:1, *c12* C18:1, *c13* C18:1, *c14* C18:1 (co-elutes with *t16* C18:1), *c15* C18:1 (co-elutes with C19:0), *c16* C18:1, *c5* C20:1, *c8* C20:1, *c11* C20:1, *c13* C22:1, *c15* C24:1

<sup>h</sup> *trans* MUFA: *t9* 14:1, *t7+t8* C16:1, *t9* C16:1, *t11+t12+t13* C16:1, *t10* C17:1, *t4* C18:1, *t5* C18:1, *t6+t7+t8* C18:1, *t9* C18:1, *t10* C18:1, VA, *t12* C18:1 co-elutes with *c6* C18:1), *t15* C18:1, *t16* C18:1 (co-elutes with *c14* C18:1)

<sup>i</sup> *cis* PUFA: LA, ALNA, *c11c14* C20:2, *c8c11c14* C20:3, *c11c14c17* C20:3, *c5c8c11c14* C20:4, *c13c16* C22:2, EPA, *c13c16c19* C22:3, DPA, DHA

<sup>j</sup> *trans* PUFA: *t11t15* C18:2, *t9t12* C18:2

<sup>k</sup> *cis/trans* + *trans/cis* PUFA: *c9t13* C18:2, *c10t14* C18:2, *c9t14* C18:2, *c9t12* C18:2, *t9c12* C18:2, *t11c15* C18:2, *t12c15* C18:2 (co-elutes with *c9* C19:1), RA (co-elutes with *t7c9+t8c10+t6c8* C18:2), other C18:2 conjugated FA of unknown isomerism

<sup>l</sup> omega-3 PUFA (n-3): *t11t15* C18:2, *t11c15* C18:2, *t12c15* C18:2 (co-elutes with *c9* C19:1), ALNA, *c11c14c17* C20:3, EPA, *c13c16c19* C22:3, DPA, DHA

<sup>m</sup> omega-6 PUFA (n-6): *t9t12* C18:2, *c9t12* C18:2, *t9c12* C18:2, LA, *c6c9c12* C18:3, *c11c14* C20:2, *c8c11c14* C20:3, *c5c8c11c14* C20:4, *c13c16* C22:2, *c7c10c13c16* C22:4

<sup>n</sup> *trans* FA: *t9* C14:1, *t9* C16:1, *t11+t12+t13* C16:1, *t10* C17:1, *t4* C18:1, *t5* C18:1, *t6+t7+t8* C18:1, *t9* C18:1, *t10* C18:1, VA, *t12* C18:1, *t15* C18:1, *t16* C18:1, *t11t15* C18:2, *t9t12* C18:2, *t12t15* C18:2

<sup>o</sup> Atherogenicity index = (C12:0 + 4 x C14:0 + C16:0) / (MUFA + PUFA), as described in Srednicka-Tober et al. (2016)

<sup>p</sup> Thrombogenicity index = (C14:0 + C16:0 + C18:0) / [(0.5 x MUFA) + (0.5 x n-6) + (3 x n-3) + (n-3/n-6)], as described in Srednicka-Tober et al. (2016)

<sup>r</sup>  $\Delta^9$ -desaturase activity index = (*c9* C14:1 + *c9* C16:1 + OA + RA) / (*c9* C14:1 + *c9* C16:1 + OA + RA + C14:0 + C16:0 + C18:0 + VA), as proposed by Kay et al. (2004)

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**Table 2**

Means (and average SE) and ANOVA P-values for the effect of month on the basic composition and fatty acid (FA) profile (g/kg total FA) of milk collected from retail outlets during the year

Parameters assessed	Month												SE	ANOVA P-values <sup>b</sup>
	March n=10	April n=10	May n=10	June n=10	July n=10	August n=10	September n=10 <sup>a</sup>	October n=10	November n=10	December n=10	January n=10	February n=10		
<b>Basic composition</b>														
Fat (g/kg milk)	38.0 <sup>AB</sup>	38.0 <sup>AB</sup>	36.8 <sup>D</sup>	36.9 <sup>CD</sup>	36.6 <sup>D</sup>	36.9 <sup>D</sup>	37.2 <sup>CD</sup>	37.2 <sup>BCD</sup>	38.5 <sup>A</sup>	36.9 <sup>D</sup>	37.7 <sup>ABC</sup>	37.7 <sup>ABC</sup>	0.49	***
Protein (g/kg milk)	32.8 <sup>BCE</sup>	32.4 <sup>CDEF</sup>	33.2 <sup>B</sup>	32.2 <sup>DEF</sup>	31.8 <sup>F</sup>	32.1 <sup>CDEF</sup>	32.4 <sup>EF</sup>	33.2 <sup>B</sup>	34.0 <sup>A</sup>	32.1 <sup>DEF</sup>	32.5 <sup>CDEF</sup>	32.1 <sup>F</sup>	0.36	***
Casein (g/kg milk)	25.2 <sup>CD</sup>	25.0 <sup>D</sup>	25.7 <sup>BC</sup>	24.8 <sup>D</sup>	24.9 <sup>D</sup>	25.3 <sup>CD</sup>	25.3 <sup>CD</sup>	26.0 <sup>AB</sup>	26.6 <sup>A</sup>	24.8 <sup>D</sup>	25.3 <sup>CD</sup>	25.0 <sup>D</sup>	0.32	***
Lactose (g/kg milk)	45.5 <sup>ABC</sup>	45.6 <sup>A</sup>	45.7 <sup>AB</sup>	45.2 <sup>BCD</sup>	44.4 <sup>EF</sup>	44.9 <sup>DE</sup>	44.2 <sup>F</sup>	44.3 <sup>F</sup>	45.4 <sup>ABCD</sup>	44.5 <sup>EF</sup>	45.2 <sup>CD</sup>	45.4 <sup>ABCD</sup>	0.19	***
SCC <sup>c</sup> (x 10 <sup>3</sup> )	52 <sup>C</sup>	48 <sup>C</sup>	49 <sup>C</sup>	30 <sup>C</sup>	40 <sup>C</sup>	38 <sup>C</sup>	44 <sup>C</sup>	38 <sup>C</sup>	35 <sup>C</sup>	219 <sup>AB</sup>	117 <sup>BC</sup>	270 <sup>A</sup>	48	***
<b>Individual FA</b>														
<b>SFA <sup>d</sup></b>														
C12:0	35.9 <sup>B</sup>	34.8 <sup>BCD</sup>	34.2 <sup>D</sup>	31.1 <sup>EF</sup>	31.4 <sup>E</sup>	31.1 <sup>EF</sup>	27.5 <sup>G</sup>	29.9 <sup>F</sup>	37.7 <sup>A</sup>	35.7 <sup>BC</sup>	35.6 <sup>BC</sup>	34.3 <sup>CD</sup>	0.61	***
C14:0	116 <sup>B</sup>	113 <sup>C</sup>	109 <sup>DE</sup>	107 <sup>E</sup>	109 <sup>D</sup>	109 <sup>DE</sup>	105 <sup>F</sup>	110 <sup>D</sup>	123 <sup>A</sup>	117 <sup>B</sup>	117 <sup>B</sup>	116 <sup>B</sup>	1.1	***
C16:0	341 <sup>A</sup>	321 <sup>C</sup>	289 <sup>E</sup>	297 <sup>D</sup>	299 <sup>D</sup>	305 <sup>D</sup>	319 <sup>C</sup>	329 <sup>B</sup>	346 <sup>A</sup>	348 <sup>A</sup>	344 <sup>A</sup>	342 <sup>A</sup>	2.7	***
C18:0	97.2 <sup>D</sup>	104.0 <sup>C</sup>	108.1 <sup>B</sup>	110.1 <sup>AB</sup>	108.7 <sup>B</sup>	107.7 <sup>B</sup>	111.3 <sup>A</sup>	104.1 <sup>C</sup>	90.4 <sup>E</sup>	96.6 <sup>D</sup>	97.3 <sup>D</sup>	99.3 <sup>D</sup>	1.80	***
<b>MUFA <sup>e</sup></b>														
VA	10.1 <sup>E</sup>	12.2 <sup>D</sup>	20.9 <sup>A</sup>	15.9 <sup>C</sup>	16.1 <sup>C</sup>	17.2 <sup>BC</sup>	18.6 <sup>B</sup>	17.7 <sup>BC</sup>	10.9 <sup>DE</sup>	9.9 <sup>E</sup>	10.2 <sup>E</sup>	10.5 <sup>DE</sup>	0.72	***
OA	186 <sup>D</sup>	195 <sup>C</sup>	209 <sup>B</sup>	212 <sup>B</sup>	211 <sup>B</sup>	211 <sup>B</sup>	223 <sup>A</sup>	213 <sup>B</sup>	183 <sup>DE</sup>	178 <sup>E</sup>	179 <sup>E</sup>	184 <sup>DE</sup>	2.5	***
<b>PUFA <sup>f</sup></b>														
LA	17.4 <sup>AB</sup>	18.1 <sup>A</sup>	17.2 <sup>ABC</sup>	16.8 <sup>BCD</sup>	15.5 <sup>EF</sup>	16.4 <sup>CDE</sup>	16.9 <sup>CD</sup>	17.2 <sup>ABC</sup>	15.7 <sup>EF</sup>	15.6 <sup>F</sup>	15.5 <sup>F</sup>	16.1 <sup>DEF</sup>	0.55	***
ALNA	4.76 <sup>E</sup>	5.65 <sup>C</sup>	8.83 <sup>A</sup>	8.31 <sup>AB</sup>	8.52 <sup>AB</sup>	8.44 <sup>AB</sup>	8.36 <sup>AB</sup>	7.97 <sup>B</sup>	5.62 <sup>CD</sup>	4.76 <sup>E</sup>	4.92 <sup>DE</sup>	4.93 <sup>DE</sup>	0.286	***
RA	4.69 <sup>D</sup>	5.20 <sup>C</sup>	6.54 <sup>A</sup>	6.09 <sup>B</sup>	6.07 <sup>B</sup>	5.99 <sup>B</sup>	6.11 <sup>B</sup>	5.51 <sup>C</sup>	4.60 <sup>D</sup>	4.52 <sup>D</sup>	4.63 <sup>D</sup>	4.72 <sup>D</sup>	0.127	***
EPA	0.502 <sup>CD</sup>	0.513 <sup>CD</sup>	0.636 <sup>A</sup>	0.641 <sup>AB</sup>	0.659 <sup>A</sup>	0.644 <sup>AB</sup>	0.647 <sup>A</sup>	0.611 <sup>B</sup>	0.535 <sup>C</sup>	0.515 <sup>CD</sup>	0.488 <sup>D</sup>	0.484 <sup>D</sup>	0.0187	***
DPA	0.786 <sup>D</sup>	0.787 <sup>D</sup>	0.878 <sup>BC</sup>	0.895 <sup>BC</sup>	0.934 <sup>AB</sup>	0.919 <sup>ABC</sup>	0.980 <sup>A</sup>	0.960 <sup>AB</sup>	0.937 <sup>AB</sup>	0.856 <sup>CD</sup>	0.918 <sup>BC</sup>	0.881 <sup>BC</sup>	0.0304	***
DHA	0.059	0.058	0.067	0.066	0.066	0.059	0.132	0.079	0.061	0.070	0.063	0.058	0.0174	ns
<b>FA groups</b>														
SFA	708 <sup>B</sup>	692 <sup>C</sup>	661 <sup>EF</sup>	665 <sup>DE</sup>	668 <sup>D</sup>	669 <sup>D</sup>	657 <sup>F</sup>	670 <sup>D</sup>	711 <sup>AB</sup>	718 <sup>A</sup>	715 <sup>AB</sup>	709 <sup>AB</sup>	3.0	***
MUFA	254 <sup>E</sup>	267 <sup>D</sup>	292 <sup>AB</sup>	290 <sup>BC</sup>	288 <sup>BC</sup>	287 <sup>BC</sup>	297 <sup>A</sup>	286 <sup>C</sup>	251 <sup>EF</sup>	246 <sup>E</sup>	248 <sup>EF</sup>	253 <sup>EF</sup>	2.7	***
<i>cis</i> MUFA <sup>g</sup>	227 <sup>D</sup>	236 <sup>C</sup>	250 <sup>B</sup>	252 <sup>B</sup>	252 <sup>B</sup>	251 <sup>B</sup>	264 <sup>A</sup>	254 <sup>B</sup>	225 <sup>D</sup>	218 <sup>E</sup>	219 <sup>E</sup>	224 <sup>DE</sup>	2.3	***
<i>trans</i> MUFA <sup>h</sup>	27.1 <sup>FG</sup>	31.1 <sup>DE</sup>	41.2 <sup>A</sup>	38.0 <sup>B</sup>	36.5 <sup>B</sup>	36.5 <sup>B</sup>	34.0 <sup>C</sup>	31.9 <sup>D</sup>	26.7 <sup>G</sup>	28.0 <sup>FG</sup>	28.8 <sup>F</sup>	29.3 <sup>EF</sup>	1.02	***
PUFA	37.9 <sup>E</sup>	41.1 <sup>D</sup>	47.2 <sup>A</sup>	44.7 <sup>BC</sup>	43.6 <sup>C</sup>	44.3 <sup>BC</sup>	45.4 <sup>B</sup>	44.3 <sup>C</sup>	37.3 <sup>EF</sup>	36.5 <sup>F</sup>	36.9 <sup>F</sup>	37.6 <sup>EF</sup>	0.51	***
<i>cis</i> PUFA <sup>i</sup>	26.1 <sup>CD</sup>	27.5 <sup>AB</sup>	28.3 <sup>A</sup>	27.3 <sup>AB</sup>	26.0 <sup>D</sup>	26.9 <sup>BC</sup>	27.8 <sup>AB</sup>	27.3 <sup>AB</sup>	24.4 <sup>F</sup>	25.1 <sup>EF</sup>	25.1 <sup>EF</sup>	25.8 <sup>DE</sup>	0.66	***
<i>trans</i> PUFA <sup>j</sup>	0.26 <sup>F</sup>	0.40 <sup>DE</sup>	0.69 <sup>A</sup>	0.49 <sup>CD</sup>	0.54 <sup>BC</sup>	0.56 <sup>BC</sup>	0.61 <sup>AB</sup>	0.52 <sup>BC</sup>	0.29 <sup>EF</sup>	0.20 <sup>F</sup>	0.26 <sup>F</sup>	0.18 <sup>F</sup>	0.047	***

<i>cis/trans</i> + <i>trans/cis</i> PUFA <sup>k</sup>	11.5 <sup>EF</sup>	13.3 <sup>C</sup>	18.2 <sup>A</sup>	16.9 <sup>B</sup>	17.0 <sup>B</sup>	16.9 <sup>B</sup>	17.1 <sup>AB</sup>	16.5 <sup>B</sup>	12.6 <sup>DE</sup>	11.2 <sup>F</sup>	11.6 <sup>EF</sup>	11.6 <sup>EF</sup>	0.55	***
n-3 <sup>l</sup>	7.74 <sup>F</sup>	8.76 <sup>D</sup>	11.71 <sup>A</sup>	10.80 <sup>B</sup>	11.01 <sup>B</sup>	10.75 <sup>B</sup>	10.99 <sup>B</sup>	10.02 <sup>C</sup>	8.10 <sup>EF</sup>	8.20 <sup>DEF</sup>	8.41 <sup>DEF</sup>	8.48 <sup>DE</sup>	0.293	***
n-6 <sup>m</sup>	20.8 <sup>AB</sup>	21.7 <sup>A</sup>	20.9 <sup>ABC</sup>	20.3 <sup>BCD</sup>	18.9 <sup>DE</sup>	19.9 <sup>BCDE</sup>	20.6 <sup>ABC</sup>	20.9 <sup>AB</sup>	19.0 <sup>E</sup>	19.4 <sup>DEF</sup>	19.3 <sup>DEF</sup>	19.9 <sup>CDEF</sup>	0.60	***
n-3/n-6	0.38 <sup>D</sup>	0.40 <sup>CD</sup>	0.57 <sup>A</sup>	0.54 <sup>A</sup>	0.59 <sup>A</sup>	0.55 <sup>A</sup>	0.55 <sup>A</sup>	0.49 <sup>B</sup>	0.43 <sup>C</sup>	0.43 <sup>B</sup>	0.44 <sup>BC</sup>	0.43 <sup>C</sup>	0.025	***
<i>trans</i> FA <sup>n</sup>	2.71 <sup>GH</sup>	3.13 <sup>DE</sup>	4.17 <sup>A</sup>	3.83 <sup>B</sup>	3.68 <sup>BC</sup>	3.69 <sup>B</sup>	3.44 <sup>C</sup>	3.23 <sup>D</sup>	2.68 <sup>H</sup>	2.84 <sup>FGH</sup>	2.93 <sup>EF</sup>	2.97 <sup>EF</sup>	0.105	***
<i>trans</i> FA (exc. VA)	1.70 <sup>D</sup>	1.92 <sup>C</sup>	2.09 <sup>B</sup>	2.24 <sup>A</sup>	2.08 <sup>B</sup>	1.97 <sup>C</sup>	1.58 <sup>E</sup>	1.46 <sup>F</sup>	1.59 <sup>E</sup>	1.85 <sup>C</sup>	1.91 <sup>C</sup>	1.92 <sup>C</sup>	0.046	***

# Indices

## Human health-related

AI <sup>o</sup>	2.90 <sup>B</sup>	2.63 <sup>C</sup>	2.24 <sup>E</sup>	2.26 <sup>E</sup>	2.32 <sup>DE</sup>	2.33 <sup>E</sup>	2.24 <sup>E</sup>	2.43 <sup>D</sup>	3.04 <sup>A</sup>	3.02 <sup>A</sup>	2.98 <sup>AB</sup>	2.89 <sup>AB</sup>	0.048	***
TI <sup>p</sup>	3.38 <sup>A</sup>	3.09 <sup>B</sup>	2.58 <sup>E</sup>	2.67 <sup>D</sup>	2.69 <sup>D</sup>	2.73 <sup>D</sup>	2.72 <sup>D</sup>	2.90 <sup>C</sup>	3.42 <sup>A</sup>	3.48 <sup>A</sup>	3.43 <sup>A</sup>	3.36 <sup>A</sup>	0.042	***

## Δ<sup>9</sup>-desaturase activity

Δ <sup>9</sup> I <sup>r</sup>	0.279 <sup>D</sup>	0.293 <sup>C</sup>	0.317 <sup>A</sup>	0.318 <sup>A</sup>	0.317 <sup>A</sup>	0.314 <sup>A</sup>	0.319 <sup>A</sup>	0.308 <sup>B</sup>	0.277 <sup>D</sup>	0.269 <sup>E</sup>	0.271 <sup>E</sup>	0.276 <sup>DE</sup>	0.0028	***
C14:1/C14:0	0.080 <sup>F</sup>	0.077 <sup>G</sup>	0.075 <sup>G</sup>	0.081 <sup>EF</sup>	0.084 <sup>C</sup>	0.084 <sup>CD</sup>	0.086 <sup>B</sup>	0.087 <sup>B</sup>	0.089 <sup>A</sup>	0.083 <sup>CD</sup>	0.082 <sup>DE</sup>	0.081 <sup>EF</sup>	0.0008	***
C16:1/C16:0	0.054 <sup>E</sup>	0.056 <sup>D</sup>	0.063 <sup>A</sup>	0.062 <sup>B</sup>	0.062 <sup>B</sup>	0.060 <sup>C</sup>	0.060 <sup>C</sup>	0.059 <sup>C</sup>	0.056 <sup>D</sup>	0.052 <sup>EF</sup>	0.052 <sup>F</sup>	0.052 <sup>F</sup>	0.0006	***
OA/C18:0	1.918 <sup>DE</sup>	1.887 <sup>EF</sup>	1.940 <sup>C</sup>	1.926 <sup>CD</sup>	1.945 <sup>CD</sup>	1.957 <sup>C</sup>	2.008 <sup>B</sup>	2.051 <sup>A</sup>	2.023 <sup>AB</sup>	1.841 <sup>G</sup>	1.838 <sup>G</sup>	1.853 <sup>FG</sup>	0.0250	***
RA/VA	0.475 <sup>BC</sup>	0.476 <sup>BC</sup>	0.433 <sup>D</sup>	0.527 <sup>A</sup>	0.537 <sup>A</sup>	0.491 <sup>B</sup>	0.449 <sup>CD</sup>	0.451 <sup>CD</sup>	0.528 <sup>A</sup>	0.483 <sup>B</sup>	0.489 <sup>B</sup>	0.474 <sup>BC</sup>	0.0113	***

<sup>a</sup> In September, there was a missing sample of free-range milk in the analysis of basic composition and the mean on this set of parameters was calculated from nine samples

<sup>b</sup> Significances were declared at \*\*\*, P < 0.001; \*\*, P < 0.01; \*, P < 0.05; †, 0.05 < P < 0.10 (trend); ns, P > 0.10 (non-significant). Means for month within a row with different upper case letters are significantly different according to Fisher's Least Significant Difference test (P < 0.05)

<sup>c</sup> Somatic cell count

<sup>d</sup> Saturated FA: C4:0, C5:0, C6:0, C7:0, C8:0, C9:0, C10:0, C11:0, C12:0, C13:0, C13:0*iso*, C13:0*anteiso*, C13:0, C14:0*iso*, C14:0, C15:0*anteiso*, C15:0, C16:0*iso*, C16:0, C17:0*iso*, C17:0, C18:0*iso*, C18:0, C20:0, C22:0, C24:0

<sup>e</sup> Monounsaturated FA: c9 C10:1, c10 C11:1, c9 C12:1, c9 C13:1, t9 14:1, c9 C14:1, c10 C15:1, t7+t8 C16:1, t9 C16:1, t11+t12+t13 C16:1, c9 C16:1 (co-elutes with C17:0*anteiso*), c11 C16:1, c13 C16:1, t10 C17:1, c9 C17:1, t4 C18:1, t5 C18:1, t6+t7+t8 C18:1, t9 C18:1, t10 C18:1, t11 C18:1 (VA), c6+t12 C18:1, c9 C18:1 (OA), t15 C18:1, c11 C18:1, c12 C18:1, c13 C18:1, t16 + c14 C18:1, c15 C18:1 (co-elutes with C19:0), c16 C18:1, c5 C20:1, c8 C20:1, c11 C20:1, c13 C22:1, c15 C24:1

<sup>f</sup> Polyunsaturated FA: t11t15 C18:2, t9t12 C18:2, c9t13 C18:2, c10t14 C18:2, c9t14 C18:2, c9t12 C18:2, t9c12 C18:2, t11c15 C18:2, c9c12 C18:2 (LA), t12c15 C18:2 (co-elutes with c9 C19:1), c6c9c12 C18:3, c9c12c15 C18:3 (ALNA), c9c11 C18:2 conjugated (RA) (co-elutes with t7c9+t8c10+t6c8 C18:2), other C18:2 conjugated FA of unknown isomerism, c11c14 C20:2, c8c11c14 C20:3, c11c14c17 C20:3, c5c8c11c14 C20:4, c13c16 C22:2, EPA, c13c16c19 C22:3, DPA, DHA

<sup>g</sup> *cis* MUFA: c9 C10:1, c10 C11:1, c9 C12:1, c9 C13:1, c9 C14:1, c10 C15:1, c9 C16:1 (co-elutes with C17:0*anteiso*), c11 C16:1, c13 C16:1, c9 C17:1, c6 C18:1 (co-elutes with t12 C18:1), OA, c11 C18:1, c12 C18:1, c13 C18:1, c14 C18:1 (co-elutes with t16 C18:1), c15 C18:1 (co-elutes with C19:0), c16 C18:1, c5 C20:1, c8 C20:1, c11 C20:1, c13 C22:1, c15 C24:1

<sup>h</sup> *trans* MUFA: t9 14:1, t7+t8 C16:1, t9 C16:1, t11+t12+t13 C16:1, t10 C17:1, t4 C18:1, t5 C18:1, t6+t7+t8 C18:1, t9 C18:1, t10 C18:1, VA, t12 C18:1 co-elutes with c6 C18:1), t15 C18:1, t16 C18:1 (co-elutes with c14 C18:1)

<sup>i</sup> *cis* PUFA: LA, ALNA, c11c14 C20:2, c8c11c14 C20:3, c11c14c17 C20:3, c5c8c11c14 C20:4, c13c16 C22:2, EPA, c13c16c19 C22:3, DPA, DHA

<sup>j</sup> *trans* PUFA: t11t15 C18:2, t9t12 C18:2

<sup>k</sup> *cis/trans* + *trans/cis* PUFA: c9t13 C18:2, c10t14 C18:2, c9t14 C18:2, c9t12 C18:2, t9c12 C18:2, t11c15 C18:2, t12c15 C18:2 (co-elutes with c9 C19:1), RA (co-elutes with t7c9+t8c10+t6c8 C18:2), other C18:2 conjugated FA of unknown isomerism

<sup>l</sup> omega-3 PUFA (n-3): t11t15 C18:2, t11c15 C18:2, t12c15 C18:2 (co-elutes with c9 C19:1), ALNA, c11c14c17 C20:3, EPA, c13c16c19 C22:3, DPA, DHA

<sup>m</sup> omega-6 PUFA (n-6): t9t12 C18:2, c9t12 C18:2, t9c12 C18:2, LA, c6c9c12 C18:3, c11c14 C20:2, c8c11c14 C20:3, c5c8c11c14 C20:4, c13c16 C22:2, c7c10c13c16 C22:4

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<sup>n</sup> *trans* FA: *t*9 C14:1, *t*9 C16:1, *t*11+*t*12+*t*13 C16:1, *t*10 C17:1, *t*4 C18:1, *t*5 C18:1, *t*6+*t*7+*t*8 C18:1, *t*9 C18:1, *t*10 C18:1, VA, *t*12 C18:1, *t*15 C18:1, *t*16 C18:1, *t*11*t*15 C18:2, *t*9*t*12 C18:2, *t*12*t*15 C18:2

<sup>o</sup> Atherogenicity index = (C12:0 + 4 x C14:0 + C16:0) / (MUFA + PUFA), as described in Srednicka-Tober et al. (2016)

<sup>p</sup> Thrombogenicity index = (C14:0 + C16:0 + C18:0) / [(0.5 x MUFA) + (0.5 x n-6) + (3 x n-3) + (n-3/n-6)], as described in Srednicka-Tober et al. (2016)

<sup>r</sup>  $\Delta^9$ -desaturase activity index = (c9 C14:1+c9 C16:1+OA+RA)/(c9 C14:1+c9 C16:1+OA+RA+C14:0+C16:0+C18:0+VA), as proposed by Kay et al. (2004)

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**Table 3**

Means (and average SE) and ANOVA P-values for the effect of production system (conventional, CON; organic, ORG; free-range, FR) on the estimated intakes of fatty acid (FA) intakes from dairy products, using milk FA profiles measured in this study.

Intakes		Males					Females					All				
Age group																
Fat (g/d) <sup>a</sup>	4-10	58.4					56.2					57.4				
	11-18	73.8					59.8					67.0				
	19-64	77.7					60.1					68.8				
	65+	74.1					57.8					65.0				
Milk fat <sup>a</sup> (% fat)	4-10	22.2					23.0					22.6				
	11-18	15.7					15.1					15.4				
	19-64	15.8					17.2					16.5				
	65+	22.4					24.4					23.5				
		Males					Females					All				
		ANOVA					ANOVA					ANOVA				
Age group	FA intakes (g/d)	CON	ORG	FR	SE	P-value <sup>b</sup>	CON	ORG	FR	SE	P-value <sup>b</sup>	CON	ORG	FR	SE	P-value <sup>b</sup>
4-10	SFA <sup>c</sup> (g/d)	8.3	8.3	8.4	0.05	ns	8.3	8.2	8.3	0.05	ns	8.3	8.3	8.4	0.05	ns
	MUFA <sup>d</sup> (g/d)	3.3	3.3	3.3	0.04	ns	3.3	3.3	3.3	0.04	ns	3.3	3.3	3.3	0.04	ns
	PUFA <sup>e</sup> (mg/d)	480 <sup>B</sup>	540 <sup>A</sup>	464 <sup>B</sup>	8.5	***	479 <sup>B</sup>	539 <sup>A</sup>	462 <sup>B</sup>	8.5	***	480 <sup>B</sup>	540 <sup>A</sup>	464 <sup>B</sup>	8.5	***
	n-3 <sup>f</sup> (mg/d)	96 <sup>B</sup>	141 <sup>A</sup>	105 <sup>B</sup>	3.3	***	96 <sup>B</sup>	141 <sup>A</sup>	105 <sup>B</sup>	3.3	***	96 <sup>B</sup>	142 <sup>A</sup>	105 <sup>B</sup>	3.3	***
	ALNA (mg/d)	53 <sup>B</sup>	81 <sup>A</sup>	58 <sup>B</sup>	1.7	***	53 <sup>B</sup>	81 <sup>A</sup>	57 <sup>B</sup>	1.7	***	53 <sup>B</sup>	81 <sup>A</sup>	58 <sup>B</sup>	1.7	***
	EPA+DHA (mg/d)	7 <sup>B</sup>	9 <sup>A</sup>	7 <sup>B</sup>	0.2	***	7 <sup>B</sup>	9 <sup>A</sup>	7 <sup>B</sup>	0.2	***	7 <sup>B</sup>	9 <sup>A</sup>	7 <sup>B</sup>	0.2	***
	trans FA <sup>g</sup> (mg/d)	378 <sup>B</sup>	429 <sup>A</sup>	367 <sup>B</sup>	10.4	***	377 <sup>B</sup>	428 <sup>A</sup>	366 <sup>B</sup>	10.4	***	378 <sup>B</sup>	430 <sup>A</sup>	367 <sup>B</sup>	10.4	***
	trans FA <sup>g</sup> (exc. VA) (mg/d)	230	223	218	5.2	ns	230	222	218	5.2	ns	230	223	218	5.2	ns
11-18	SFA <sup>c</sup> (g/d)	7.4	7.4	7.5	0.05	ns	5.8	5.8	5.8	0.04	ns	6.6	6.6	6.7	0.04	ns
	MUFA <sup>d</sup> (g/d)	2.9	2.9	2.9	0.04	ns	2.3	2.3	2.3	0.03	ns	2.6	2.6	2.6	0.03	ns
	PUFA <sup>e</sup> (mg/d)	429 <sup>B</sup>	483 <sup>A</sup>	414 <sup>B</sup>	7.6	***	334 <sup>B</sup>	376 <sup>A</sup>	323 <sup>B</sup>	5.9	***	382 <sup>B</sup>	430 <sup>A</sup>	369 <sup>B</sup>	6.8	***
	n-3 <sup>f</sup> (mg/d)	86 <sup>B</sup>	126 <sup>A</sup>	94 <sup>B</sup>	3.0	***	67 <sup>B</sup>	99 <sup>A</sup>	73 <sup>B</sup>	2.3	***	76 <sup>B</sup>	113 <sup>A</sup>	83 <sup>B</sup>	2.6	***
	ALNA (mg/d)	47 <sup>B</sup>	73 <sup>A</sup>	51 <sup>B</sup>	1.5	***	37 <sup>B</sup>	57 <sup>A</sup>	40 <sup>B</sup>	1.2	***	42 <sup>B</sup>	65 <sup>A</sup>	46 <sup>B</sup>	1.4	***
	EPA+DHA (mg/d)	6 <sup>B</sup>	8 <sup>A</sup>	7 <sup>B</sup>	0.2	***	5 <sup>B</sup>	6 <sup>A</sup>	5 <sup>B</sup>	0.1	***	5 <sup>B</sup>	7 <sup>A</sup>	6 <sup>B</sup>	0.2	***
	trans FA <sup>g</sup> (mg/d)	338 <sup>B</sup>	384 <sup>A</sup>	328 <sup>B</sup>	9.3	***	263 <sup>B</sup>	299 <sup>A</sup>	255 <sup>B</sup>	7.2	***	301 <sup>B</sup>	342 <sup>A</sup>	292 <sup>B</sup>	8.3	***
	trans FA <sup>g</sup> (exc. VA) (mg/d)	206	199	195	4.7	ns	160	155	152	3.6	ns	183	177	174	4.2	ns
19-64	SFA <sup>c</sup> (g/d)	7.9	7.8	7.9	0.05	ns	6.6	6.6	6.7	0.04	ns	7.3	7.2	7.3	0.05	ns
	MUFA <sup>d</sup> (g/d)	3.1	3.1	3.1	0.04	ns	2.6	2.6	2.6	0.03	ns	2.9	2.9	2.9	0.04	ns
	PUFA <sup>e</sup> (mg/d)	455 <sup>B</sup>	511 <sup>A</sup>	439 <sup>B</sup>	8.0	***	383 <sup>B</sup>	431 <sup>A</sup>	370 <sup>B</sup>	6.8	***	420 <sup>B</sup>	473 <sup>A</sup>	406 <sup>B</sup>	7.4	***
	n-3 <sup>f</sup> (mg/d)	91 <sup>B</sup>	134 <sup>A</sup>	99 <sup>B</sup>	3.1	***	76 <sup>B</sup>	113 <sup>A</sup>	84 <sup>B</sup>	2.6	***	84 <sup>B</sup>	124 <sup>A</sup>	92 <sup>B</sup>	2.9	***
	ALNA	50 <sup>B</sup>	77 <sup>A</sup>	55 <sup>B</sup>	1.6	***	42 <sup>B</sup>	65 <sup>A</sup>	46 <sup>B</sup>	1.4	***	46 <sup>B</sup>	71 <sup>A</sup>	50 <sup>B</sup>	1.5	***

65+	EPA+DHA	6 <sup>B</sup>	9 <sup>A</sup>	7 <sup>B</sup>	0.2	***	5 <sup>B</sup>	7 <sup>A</sup>	6 <sup>B</sup>	0.2	***	6 <sup>B</sup>	8 <sup>A</sup>	6 <sup>B</sup>	0.2	***
	<i>trans</i> FA <sup>g</sup> (mg/d)	358 <sup>B</sup>	407 <sup>A</sup>	347 <sup>B</sup>	9.8	***	301 <sup>B</sup>	342 <sup>A</sup>	292 <sup>B</sup>	8.3	***	331 <sup>B</sup>	376 <sup>A</sup>	321 <sup>B</sup>	9.1	***
	<i>trans</i> <sup>g</sup> FA (exc. VA) (mg/d)	218	211	207	5.0	ns	184	178	174	4.2	ns	202	195	191	4.6	ns
	SFA <sup>c</sup> (g/d)	10.7	10.6	10.7	0.07	ns	9.0	9.0	9.1	0.06	ns	9.8	9.7	9.9	0.06	ns
	MUFA <sup>d</sup> (g/d)	4.2	4.2	4.2	0.06	ns	3.6	3.6	3.5	0.05	ns	3.9	3.9	3.8	0.05	ns
	PUFA <sup>e</sup> (mg/d)	615 <sup>B</sup>	692 <sup>A</sup>	593 <sup>B</sup>	10.9	***	522 <sup>B</sup>	588 <sup>A</sup>	504 <sup>B</sup>	9.2	***	566 <sup>B</sup>	636 <sup>A</sup>	546 <sup>B</sup>	10.0	***
	n-3 <sup>f</sup> (mg/d)	123 <sup>B</sup>	181 <sup>A</sup>	134 <sup>B</sup>	4.2	***	104 <sup>B</sup>	154 <sup>A</sup>	114 <sup>B</sup>	3.6	***	113 <sup>B</sup>	167 <sup>A</sup>	124 <sup>B</sup>	3.9	***
	ALNA	68 <sup>B</sup>	104 <sup>A</sup>	74 <sup>B</sup>	2.2	***	58 <sup>B</sup>	88 <sup>A</sup>	63 <sup>B</sup>	1.9	***	63 <sup>B</sup>	96 <sup>A</sup>	68 <sup>B</sup>	2.0	***
	EPA+DHA	9 <sup>B</sup>	12 <sup>A</sup>	9 <sup>B</sup>	0.3	***	7 <sup>B</sup>	10 <sup>A</sup>	8 <sup>B</sup>	0.2	***	8 <sup>B</sup>	11 <sup>A</sup>	9 <sup>B</sup>	0.2	***
	<i>trans</i> FA <sup>g</sup> (mg/d)	484 <sup>B</sup>	550 <sup>A</sup>	469 <sup>B</sup>	13.3	***	411 <sup>B</sup>	467 <sup>A</sup>	399 <sup>B</sup>	11.3	***	445 <sup>B</sup>	506 <sup>A</sup>	432 <sup>B</sup>	12.3	***
	<i>trans</i> <sup>g</sup> FA (exc. VA) (mg/d)	295	285	280	6.7	ns	251	242	238	5.7	ns	271	262	257	6.2	ns

<sup>a</sup> Intake data of fats and fatty acids of Year 1 of the National Diet and Nutrition Survey rolling programme 2008-2009, as presented by Bates et al. (2014). For the purposes of the intake calculations, this study assumes that all dairy products produced in the UK have the same FA profile as the whole milk analysed.

<sup>b</sup> Significances were declared at \*\*\*,  $P < 0.001$ ; \*\*,  $P < 0.01$ ; \*,  $P < 0.05$ ; †,  $0.05 < P < 0.10$  (trend); ns,  $P > 0.10$  (non-significant). Means for production system within a row and gender with different upper case letters are significantly different according to Fisher's Least Significant Difference test ( $P < 0.05$ )

<sup>d</sup> Saturated FA: C4:0, C5:0, C6:0, C7:0, C8:0, C9:0, C10:0, C11:0, C12:0, C13:0, C13:0*iso*, C13:0*anteiso*, C13:0, C14:0*iso*, C14:0, C15:0*anteiso*, C15:0, C16:0*iso*, C16:0, C17:0*iso*, C17:0, C18:0*iso*, C18:0, C20:0, C22:0, C24:0

<sup>e</sup> Monounsaturated FA: *c*9 C10:1, *c*10 C11:1, *c*9 C12:1, *c*9 C13:1, *t*9 14:1, *c*9 C14:1, *c*10 C15:1, *t*7+*t*8 C16:1, *t*9 C16:1, *t*11+*t*12+*t*13 C16:1, *c*9 C16:1 (co-elutes with C17:0*anteiso*), *c*11 C16:1, *c*13 C16:1, *t*10 C17:1, *c*9 C17:1, *t*4 C18:1, *t*5 C18:1, *t*6+*t*7+*t*8 C18:1, *t*9 C18:1, *t*10 C18:1, *t*11 C18:1 (VA), *c*6+*t*12 C18:1, *c*9 C18:1 (OA), *t*15 C18:1, *c*11 C18:1, *c*12 C18:1, *c*13 C18:1, *t*16 + *c*14 C18:1, *c*15 C18:1 (co-elutes with C19:0), *c*16 C18:1, *c*5 C20:1, *c*8 C20:1, *c*11 C20:1, *c*13 C22:1, *c*15 C24:1

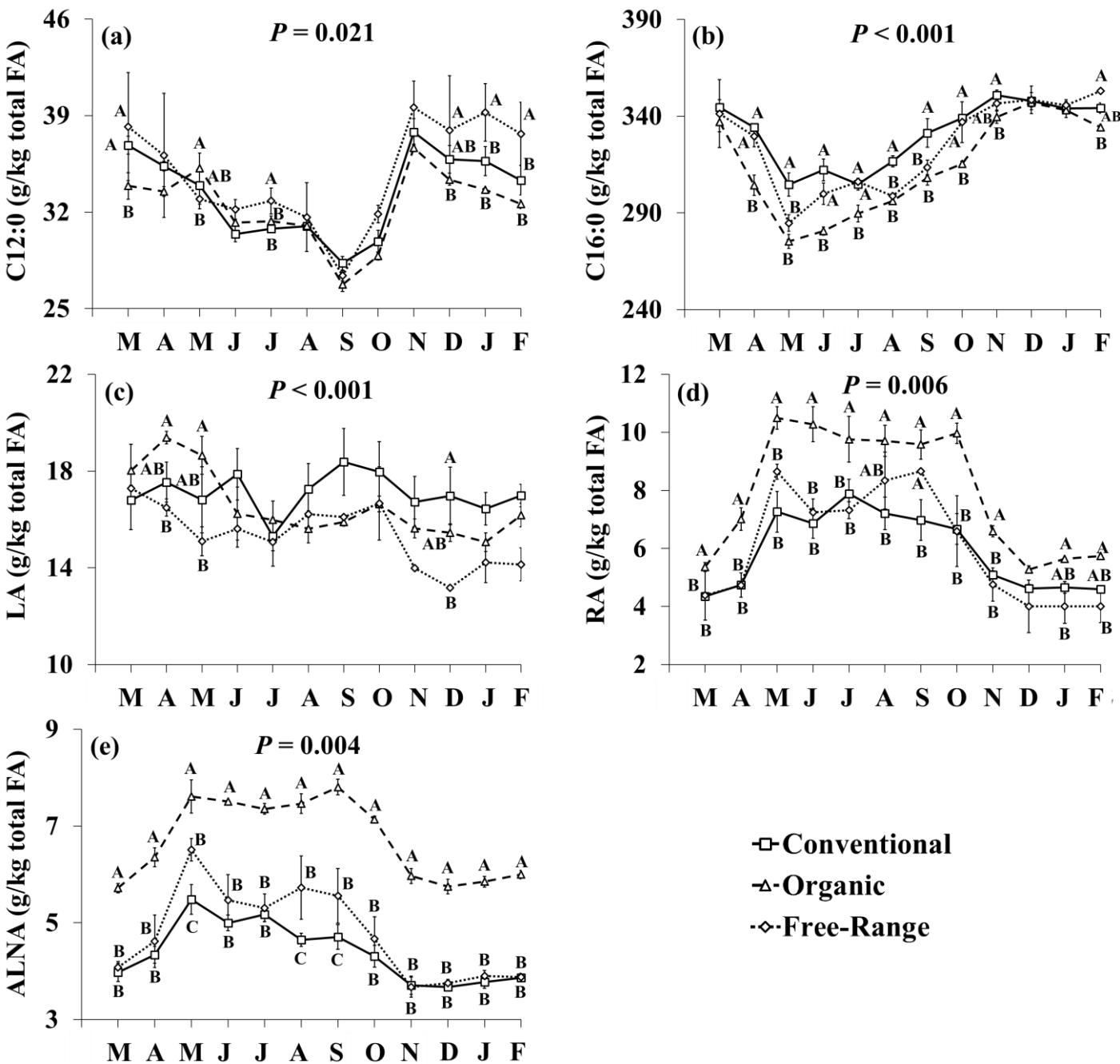
<sup>f</sup> Polyunsaturated FA: *t*11*t*15 C18:2, *t*9*t*12 C18:2, *c*9*t*13 C18:2, *c*10*t*14 C18:2, *c*9*t*14 C18:2, *c*9*t*12 C18:2, *t*9*c*12 C18:2, *t*11*c*15 C18:2, *c*9*c*12 C18:2 (LA), *t*12*c*15 C18:2 (co-elutes with *c*9 C19:1), *c*6*c*9*c*12 C18:3, *c*9*c*12*c*15 C18:3 (ALNA), *c*9*c*11 C18:2 conjugated (RA) (co-elutes with *t*7*c*9+*t*8*c*10+*t*6*c*8 C18:2), other C18:2 conjugated FA of unknown isomerism, *c*11*c*14 C20:2, *c*8*c*11*c*14 C20:3, *c*11*c*14*c*17 C20:3, *c*5*c*8*c*11*c*14 C20:4, *c*13*c*16 C22:2, EPA, *c*13*c*16*c*19 C22:3, DPA, DHA

<sup>g</sup> omega-3 PUFA (n-6): *t*11*t*15 C18:2, *t*11*c*15 C18:2, *t*12*c*15 C18:2 (co-elutes with *c*9 C19:1), ALNA, *c*11*c*14*c*17 C20:3, EPA, *c*13*c*16*c*19 C22:3, DPA, DHA

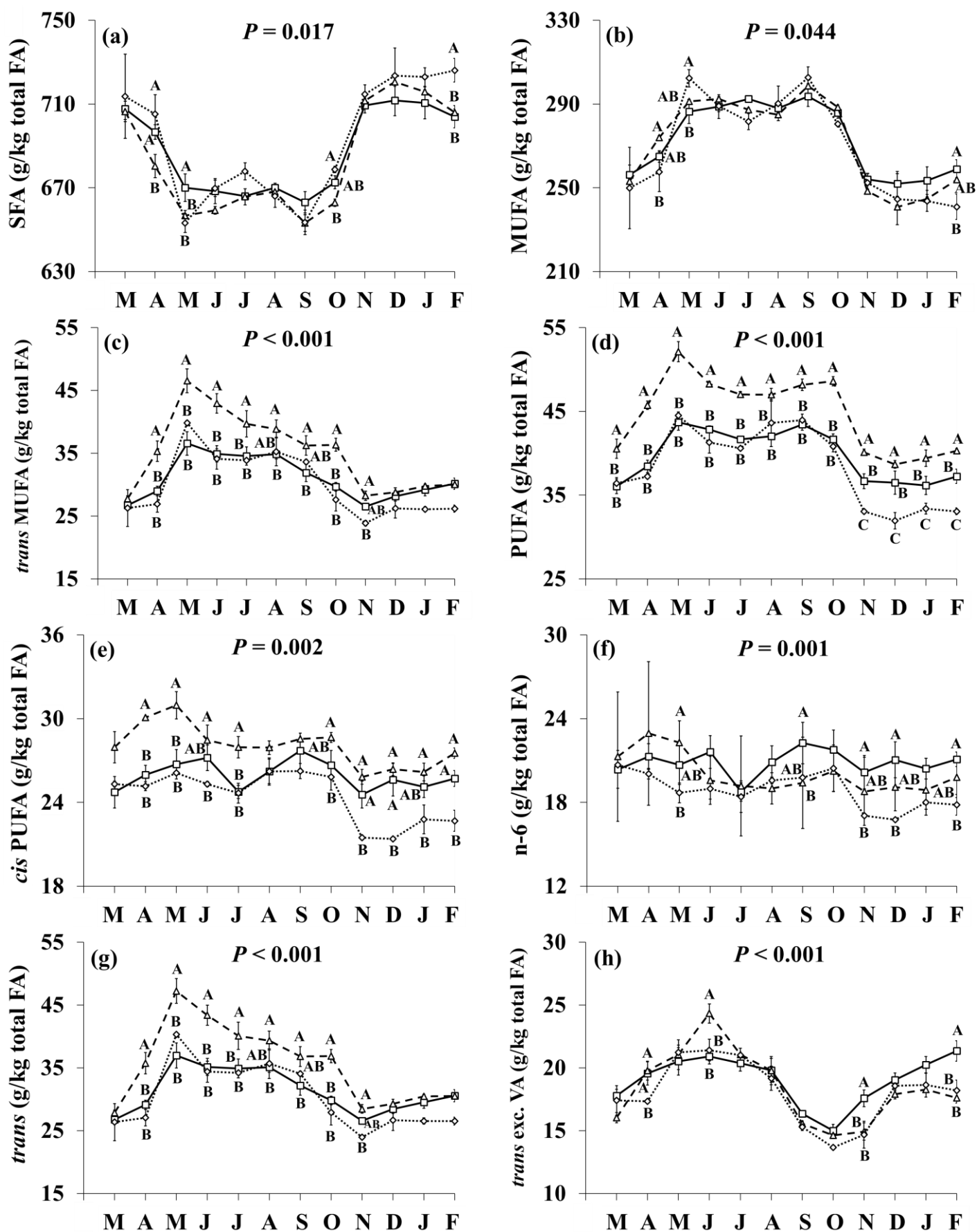
<sup>h</sup> omega-6 PUFA (n-6): *t*9*t*12 C18:2, *c*9*t*12 C18:2, *t*9*c*12 C18:2, LA, *c*6*c*9*c*12 C18:3, *c*11*c*14 C20:2, *c*8*c*11*c*14 C20:3, *c*5*c*8*c*11*c*14 C20:4, *c*13*c*16 C22:2, *c*7*c*10*c*13*c*16 C22:4

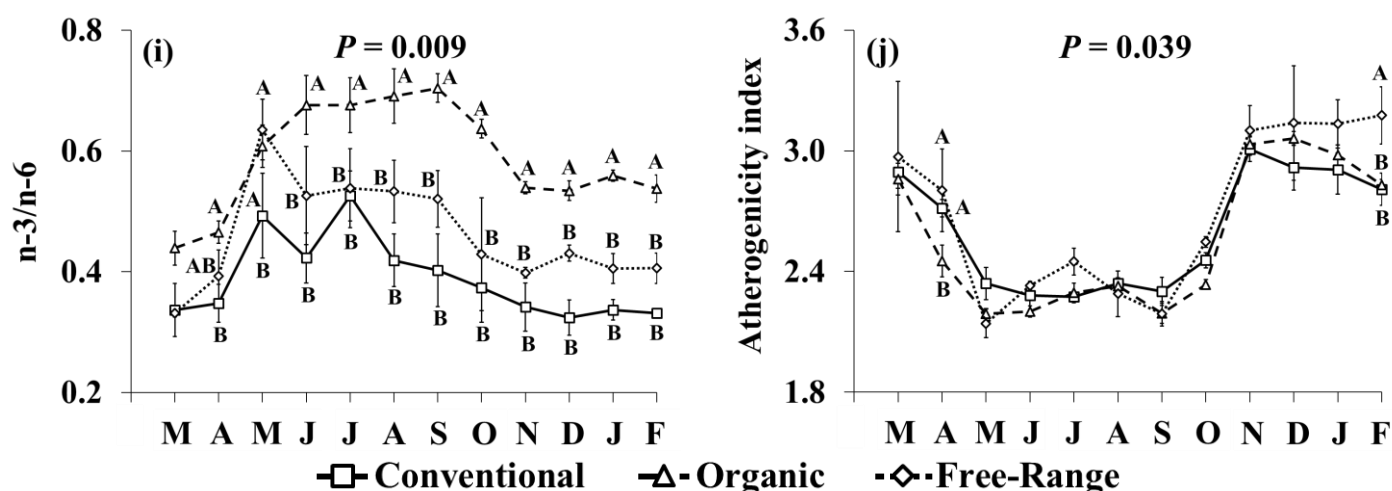
<sup>i</sup> *trans* FA: *t*9 C14:1, *t*9 C16:1, *t*11+*t*12+*t*13 C16:1, *t*10 C17:1, *t*4 C18:1, *t*5 C18:1, *t*6+*t*7+*t*8 C18:1, *t*9 C18:1, *t*10 C18:1, VA, *t*12 C18:1, *t*15 C18:1, *t*16 C18:1, *t*11*t*15 C18:2, *t*9*t*12 C18:2, *t*12*t*15 C18:2



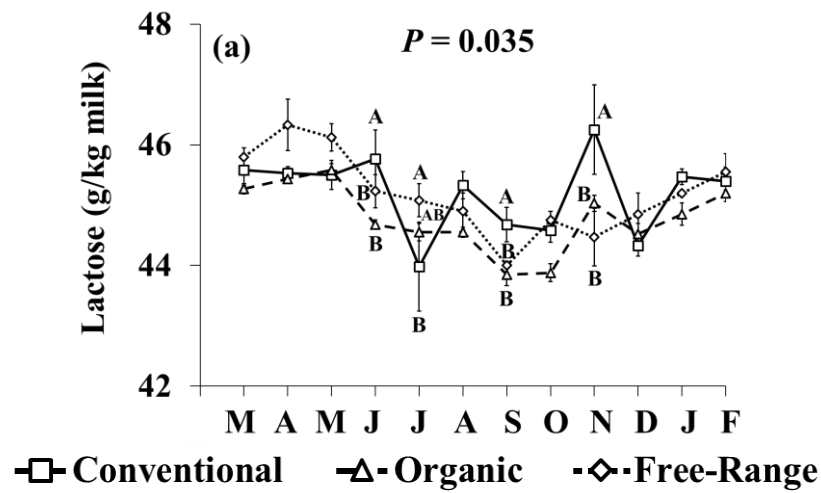


**Figure 1.** Interaction means  $\pm$  SE (error bars) for the effects of production system (conventional, CON; organic, ORG; free-range, FR) and month (in order of appearance from left to right in Axis Y: M, March; A, April; M, May; J, June; J, July; A, August; S, September; O, October; N, November; D, December; J, January; F, February) on the concentrations of (a) C12:0, (b) C16:0, (c) linoleic acid (LA), (d) rumenic acid (RA) and (e)  $\alpha$ -linolenic acid (ALNA) of milk collected from retail outlets during the year. P represents the ANOVA P-value for the interaction. Means for production system and within a month with different upper case letters are significantly different according to Fisher's Least Significant Difference test ( $P < 0.05$ ).

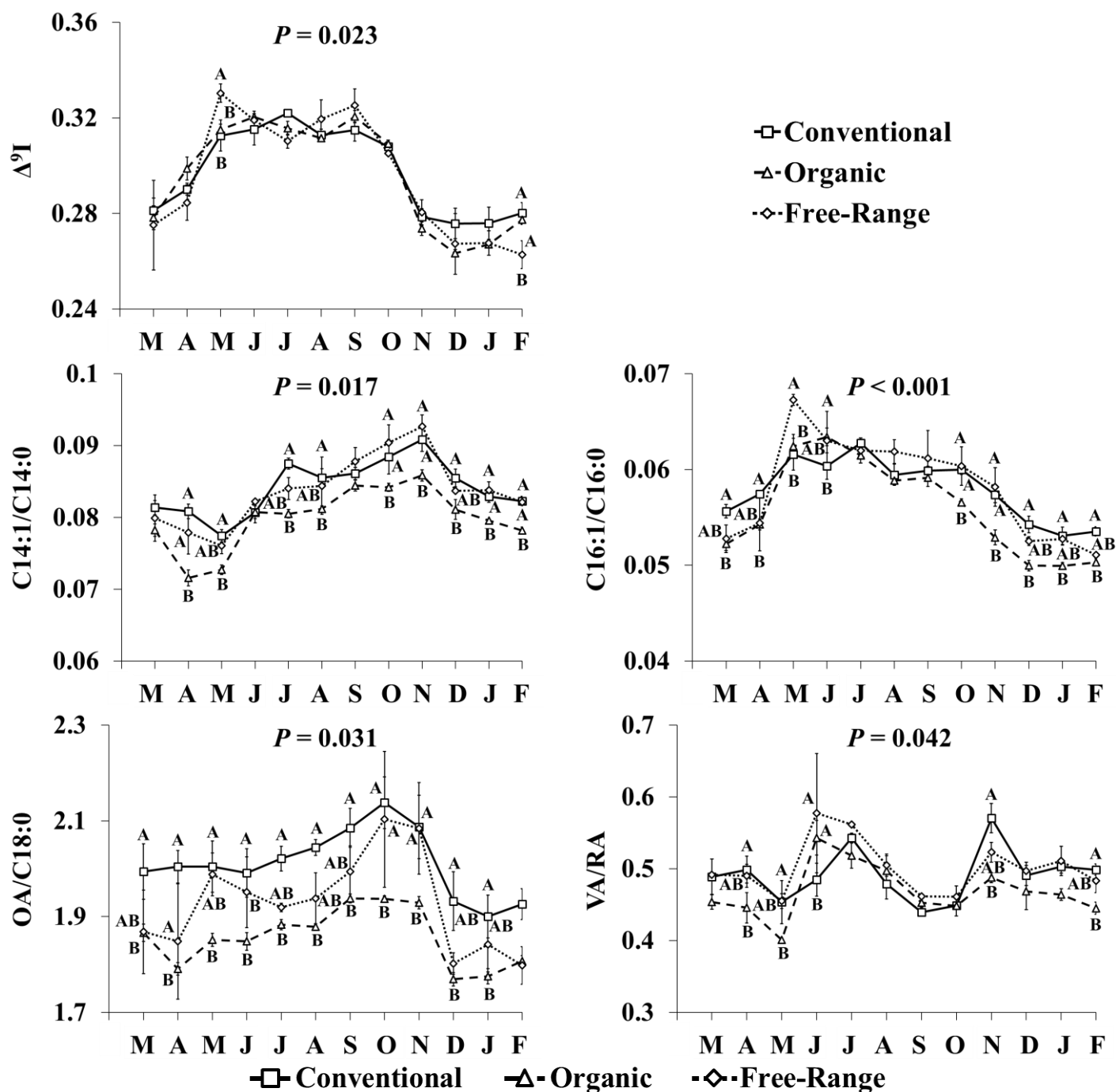




**Figure 2.** Interaction means  $\pm$  SE (error bars) for the effects of production system (conventional, CON; organic, ORG; free-range, FR) and month (in order of appearance from left to right in Axis Y: M, March; A, April; M, May; J, June; J, July; A, August; S, September; O, October; N, November; D, December; J, January; F, February) on the concentrations of (a) saturated fatty acids (SFA), (b) monounsaturated fatty acids (MUFA), (c) *trans* MUFA, (d) polyunsaturated fatty acids (PUFA), (e) *cis* PUFA, (f) omega-6 PUFA (n-6), (g) *trans* FA, *trans* FA (h) excluding VA, (i) the ratio of omega-3 PUFA/omega-6 PUFA (n-3/n-6) and (j) the atherogenicity index (as proposed by Srednicka-Tober et al. (2016)) of milk collected from retail outlets during the year. P represents the ANOVA P-value for the interaction. Means for production system and within a month with different upper case letters are significantly different according to Fisher's Least Significant Difference test ( $P < 0.05$ ).



**Figure A1.** Interaction means  $\pm$  SE (error bars) for the effects of production system (conventional, organic, free-range) and month (in order of appearance from left to right in Axis Y: M, March; A, April; M, May; J, June; J, July; A, August; S, September; O, October; N, November; D, December; J, January; F, February) on the concentrations of lactose of milk collected from retail outlets during the year. P represents the ANOVA P-value for the interaction. Means for production system and within a month with different upper case letters are significantly different according to Fisher's Least Significant Difference test ( $P < 0.05$ ).



**Figure A2.** Interaction means  $\pm$  SE (error bars) for the effects of production system (conventional, organic, free-range) and month (in order of appearance from left to right in Axis Y: M, March; A, April; M, May; J, June; J, July; A, August; S, September; O, October; N, November; D, December; J, January; F, February) on the  $\Delta^9$ -desaturase activity indices ( $\Delta^9I$  as proposed by Kay et al. (2004), and ratios of C14:1/C14:0, C16:1/C16:0, oleic acid (OA)/C18:0 and vaccenic acid (VA)/ruminic acid (RA)) of milk collected from retail outlets during the year. P represents the ANOVA P-value for the interaction. Means for production system and within a month with different upper case letters are significantly different according to Fisher's Least Significant Difference test ( $P < 0.05$ ).

**Table A1**

Means (and average SE) and ANOVA P-values for the effect of production system (conventional, CON; organic, ORG; free-range, FR) on the fatty acid profile (g/kg total fatty acids) of milk collected from retail outlets during the year

Parameters assessed	Production System			SE	ANOVA P-values <sup>b</sup>
	CON n=48	ORG n=48	FR n=24 <sup>a</sup>		
C4:0	20.7	21.2	20.9	0.54	†
C5:0	0.207	0.230	0.194	0.0189	ns
C6:0	15.3 <sup>B</sup>	15.9 <sup>A</sup>	15.5 <sup>AB</sup>	0.38	*
C7:0	0.218 <sup>A</sup>	0.174 <sup>B</sup>	0.206 <sup>AB</sup>	0.0123	*
C8:0	10.0	10.3	10.1	0.23	ns
C9:0	0.288	0.244	0.270	0.0122	ns
C10:0	25.1	25.7	25.5	0.55	ns
c9 C10:1	2.44	2.55	2.52	0.045	ns
C11:0	0.547	0.459	0.509	0.0228	ns
C12:0	33.4	32.4	34.8	0.60	ns
C13:0 iso	0.271 <sup>B</sup>	0.334 <sup>A</sup>	0.291 <sup>B</sup>	0.0104	**
C13:0 anteiso	0.145	0.110	0.116	0.0087	†
c9 C12:1	0.830	0.799	0.861	0.0173	ns
C13:0	0.908	0.870	0.848	0.0265	ns
C14:0 iso	0.812 <sup>B</sup>	1.037 <sup>A</sup>	0.836 <sup>B</sup>	0.0180	***
C14:0	111	114	114	1.1	ns
t9 C14:1	2.16 <sup>C</sup>	2.56 <sup>A</sup>	2.37 <sup>B</sup>	0.042	***
C15:0 anteiso	4.23 <sup>B</sup>	4.74 <sup>A</sup>	4.65 <sup>A</sup>	0.098	**
c9 C14:1	9.31	9.09	9.56	0.138	ns
C15:0	10.3 <sup>B</sup>	11.1 <sup>A</sup>	10.6 <sup>B</sup>	0.13	**
C16:0 iso	1.99 <sup>B</sup>	2.26 <sup>A</sup>	1.97 <sup>B</sup>	0.028	***
C16:0	331 <sup>A</sup>	314 <sup>B</sup>	325 <sup>A</sup>	3.7	***
t6+t7+t8 C16:1	0.316 <sup>A</sup>	0.293 <sup>B</sup>	0.290 <sup>B</sup>	0.0071	*
t9 C16:1	0.092	0.081	0.083	0.0030	ns
C17:0 iso	3.84 <sup>B</sup>	4.28 <sup>A</sup>	4.09 <sup>AB</sup>	0.110	*
t11+t12+t13 C16:1	1.96	1.87	1.92	0.047	ns
c9 C16:1 + C17 anteiso	19.1 <sup>A</sup>	17.5 <sup>B</sup>	18.8 <sup>A</sup>	0.14	***
c11 C16:1	1.43	1.48	1.89	0.053	†
c13 C16:1	1.40	1.34	1.40	0.052	ns
C17:0	4.87 <sup>B</sup>	5.69 <sup>A</sup>	5.02 <sup>B</sup>	0.070	***
t10 c17:1	0.501 <sup>B</sup>	0.551 <sup>A</sup>	0.477 <sup>C</sup>	0.0138	***
C18:0 iso	0.571	0.636	0.706	0.0195	ns
c9 C17:1	2.11 <sup>B</sup>	2.27 <sup>A</sup>	2.15 <sup>AB</sup>	0.035	*
C18:0	100	106	103	1.4	ns
t4 C18:1	0.158 <sup>A</sup>	0.128 <sup>B</sup>	0.120 <sup>B</sup>	0.0049	**
t5 C18:1	0.111 <sup>A</sup>	0.088 <sup>B</sup>	0.092 <sup>AB</sup>	0.0039	*
t6+t7+t8 C18:1	2.76	2.55	2.40	0.048	†
t9 C18:1	1.93 <sup>A</sup>	1.66 <sup>B</sup>	1.72 <sup>AB</sup>	0.045	*
t10 C18:1	3.70	3.49	3.40	0.241	ns
t11 C18:1	12.2 <sup>B</sup>	17.1 <sup>A</sup>	12.3 <sup>B</sup>	0.71	***
c6 + t12 C18:1	2.83 <sup>A</sup>	2.30 <sup>B</sup>	2.50 <sup>B</sup>	0.124	**
c9 C18:1	200	197	199	3.0	ns
t15 C18:1	2.08 <sup>A</sup>	1.89 <sup>B</sup>	1.97 <sup>AB</sup>	0.146	*
c11 C18:1	5.89 <sup>A</sup>	4.61 <sup>B</sup>	4.90 <sup>B</sup>	0.099	*
c12 C18:1	2.24	1.94	1.90	0.057	ns
c13 C18:1	0.913 <sup>A</sup>	0.819 <sup>B</sup>	0.864 <sup>AB</sup>	0.0160	*
t16 + c14 C18:1	3.25	3.29	3.27	0.049	ns
c15 C18:1 + C19:0	1.27	1.35	1.34	0.059	†
t11t15 C18:2	0.269 <sup>B</sup>	0.444 <sup>A</sup>	0.314 <sup>B</sup>	0.0278	*
t9t12 C18:2	0.071	0.072	0.058	0.0072	ns

c9t13 C18:2	2.10	2.00	1.99	0.052	ns
c10t14 C18:2	1.10	1.08	1.00	0.024	ns
c9t14 C18:2	1.25 <sup>A</sup>	1.14 <sup>B</sup>	1.18 <sup>AB</sup>	0.022	*
c9t12 C18:2	0.620 <sup>A</sup>	0.596 <sup>B</sup>	0.614 <sup>AB</sup>	0.0087	*
c16 C18:1	0.336	0.371	0.320	0.0166	ns
t11c15 C18:2	1.50 <sup>B</sup>	2.23 <sup>A</sup>	1.71 <sup>B</sup>	0.101	**
t9c12 C18:2	0.194 <sup>A</sup>	0.118 <sup>C</sup>	0.157 <sup>B</sup>	0.0125	***
c9c12 C18:2	17.1	16.6	15.3	0.28	ns
t12c15 C18:2 + c9 C19:1	0.430	0.480	0.480	0.0461	ns
C20:0	1.40 <sup>B</sup>	1.60 <sup>A</sup>	1.39 <sup>B</sup>	0.025	**
c6c9c12 C18:3	0.258	0.229	0.226	0.0069	ns
c8 C20:1	1.02 <sup>B</sup>	1.13 <sup>A</sup>	1.01 <sup>B</sup>	0.014	**
c11 C20:1	0.393	0.346	0.347	0.0295	†
c9c12c15 C18:3	4.39 <sup>B</sup>	6.71 <sup>A</sup>	4.76 <sup>B</sup>	0.141	***
c9t11 C18:2	5.91 <sup>B</sup>	7.95 <sup>A</sup>	6.06 <sup>B</sup>	0.315	***
Unknown C18:2 conjugated	0.286 <sup>B</sup>	0.344 <sup>A</sup>	0.277 <sup>B</sup>	0.0172	*
Unknown C18:2 conjugated	0.265 <sup>B</sup>	0.413 <sup>A</sup>	0.288 <sup>B</sup>	0.0134	***
c11c14 C20:2	0.196 <sup>B</sup>	0.220 <sup>A</sup>	0.196 <sup>B</sup>	0.0156	*
C22:0	0.556 <sup>B</sup>	0.735 <sup>A</sup>	0.578 <sup>B</sup>	0.0118	***
c8c11c14 C20:3	0.809	0.704	0.713	0.0105	†
c13 C22:1	0.160	0.182	0.093	0.0129	ns
c11c14c17 C20:3	0.105 <sup>B</sup>	0.186 <sup>A</sup>	0.102 <sup>B</sup>	0.0102	***
c5c8c11c14 C20:4	1.06	0.95	0.94	0.016	†
c13c16 C22:2	0.397	0.452	0.495	0.0138	ns
c5c8c11c14c17 C20:5	0.484 <sup>B</sup>	0.674 <sup>A</sup>	0.551 <sup>B</sup>	0.0143	***
C24:0	0.345 <sup>B</sup>	0.457 <sup>A</sup>	0.362 <sup>B</sup>	0.0063	***
c13c16c19 C22:3	0.104	0.114	0.096	0.0070	†
c7c10c13c16 C22:4	0.170 <sup>A</sup>	0.132 <sup>B</sup>	0.114 <sup>B</sup>	0.0113	*
c7c10c13c16c19 C22:5	0.795 <sup>B</sup>	1.024 <sup>A</sup>	0.834 <sup>B</sup>	0.0161	***
c4c7c10c13c16c19 C22:6	0.067	0.078	0.060	0.0051	ns

<sup>a</sup> In September, there was a missing sample of free-range milk in the analysis of basic composition and the mean on this set of parameters was calculated from 23 samples

<sup>b</sup> Significances were declared at \*\*\*,  $P < 0.001$ ; \*\*,  $P < 0.01$ ; \*,  $P < 0.05$ ; †,  $0.05 < P < 0.10$  (trend); ns,  $P > 0.10$  (non-significant). Means for production system within a row with different upper case letters are significantly different according to Fisher's Least Significant Difference test ( $P < 0.05$ )

**Table A2**

Means (and average SE) and ANOVA P-values for the effect of month on the fatty acid profile (g/kg total fatty acids) of milk collected from retail outlets during the year

Parameters assessed	Month												SE	ANOVA P-values <sup>b</sup>
	March n=10	April n=10	May n=10	June n=10	July n=10	August n=10	September n=10 <sup>a</sup>	October n=10	November n=10	December n=10	January n=10	February n=10		
C4:0	20.8 <sup>D</sup>	22.4 <sup>BC</sup>	22.7 <sup>B</sup>	23.2 <sup>A</sup>	23.5 <sup>A</sup>	22.4 <sup>BC</sup>	14.7 <sup>F</sup>	14.3 <sup>F</sup>	18.3 <sup>E</sup>	23.3 <sup>A</sup>	23.6 <sup>A</sup>	22.0 <sup>C</sup>	0.19	***
C5:0	0.224	0.215	0.215	0.174	0.169	0.172	0.125	0.140	0.202	0.235	0.296	0.396	0.0258	ns
C6:0	16.5 <sup>BC</sup>	16.7 <sup>B</sup>	16.5 <sup>C</sup>	16.6 <sup>BC</sup>	16.5 <sup>C</sup>	16.1 <sup>D</sup>	10.8 <sup>F</sup>	10.9 <sup>F</sup>	15.1 <sup>E</sup>	17.3 <sup>A</sup>	17.4 <sup>A</sup>	16.7 <sup>B</sup>	0.14	***
C7:0	0.231 <sup>BC</sup>	0.223 <sup>BC</sup>	0.219 <sup>C</sup>	0.163 <sup>D</sup>	0.151 <sup>DE</sup>	0.155 <sup>D</sup>	0.115 <sup>E</sup>	0.141 <sup>DE</sup>	0.208 <sup>C</sup>	0.245 <sup>BC</sup>	0.274 <sup>A</sup>	0.253 <sup>B</sup>	0.0136	***
C8:0	10.9 <sup>AB</sup>	11.0 <sup>AB</sup>	11.0 <sup>AB</sup>	10.6 <sup>CD</sup>	10.4 <sup>DE</sup>	10.2 <sup>E</sup>	7.1 <sup>G</sup>	7.4 <sup>F</sup>	10.4 <sup>DE</sup>	11.1 <sup>A</sup>	11.0 <sup>A</sup>	10.7 <sup>BC</sup>	0.13	***
C9:0	0.317 <sup>AB</sup>	0.303 <sup>B</sup>	0.311 <sup>B</sup>	0.217 <sup>C</sup>	0.210 <sup>C</sup>	0.215 <sup>C</sup>	0.173 <sup>D</sup>	0.208 <sup>C</sup>	0.308 <sup>AB</sup>	0.318 <sup>AB</sup>	0.326 <sup>A</sup>	0.297 <sup>B</sup>	0.0138	***
C10:0	27.6 <sup>A</sup>	27.5 <sup>AB</sup>	27.5 <sup>AB</sup>	25.3 <sup>C</sup>	24.9 <sup>C</sup>	24.5 <sup>C</sup>	18.9 <sup>E</sup>	20.3 <sup>D</sup>	27.7 <sup>A</sup>	27.3 <sup>AB</sup>	27.2 <sup>AB</sup>	26.4 <sup>B</sup>	0.44	***
c9 C10:1	2.62 <sup>BCD</sup>	2.50 <sup>EF</sup>	2.43 <sup>G</sup>	2.46 <sup>FG</sup>	2.56 <sup>DEF</sup>	2.54 <sup>DEF</sup>	1.99 <sup>I</sup>	2.10 <sup>H</sup>	2.85 <sup>A</sup>	2.69 <sup>B</sup>	2.65 <sup>BC</sup>	2.58 <sup>CDE</sup>	0.041	***
C11:0	0.587 <sup>A</sup>	0.552 <sup>A</sup>	0.584 <sup>A</sup>	0.394 <sup>BC</sup>	0.381 <sup>C</sup>	0.391 <sup>BC</sup>	0.348 <sup>C</sup>	0.429 <sup>B</sup>	0.607 <sup>A</sup>	0.610 <sup>A</sup>	0.594 <sup>A</sup>	0.573 <sup>A</sup>	0.0267	***
C12:0	35.9 <sup>B</sup>	34.8 <sup>BCD</sup>	34.2 <sup>D</sup>	31.1 <sup>EF</sup>	31.4 <sup>E</sup>	31.1 <sup>EF</sup>	27.5 <sup>G</sup>	29.9 <sup>F</sup>	37.7 <sup>A</sup>	35.7 <sup>BC</sup>	35.6 <sup>BC</sup>	34.3 <sup>CD</sup>	0.62	***
C13:0 iso	0.280 <sup>E</sup>	0.280 <sup>E</sup>	0.306 <sup>C</sup>	0.377 <sup>A</sup>	0.380 <sup>A</sup>	0.380 <sup>A</sup>	0.332 <sup>B</sup>	0.300 <sup>CD</sup>	0.288 <sup>DE</sup>	0.221 <sup>F</sup>	0.229 <sup>F</sup>	0.227 <sup>F</sup>	0.0125	***
C13:0 anteiso	0.113 <sup>B</sup>	0.105 <sup>B</sup>	0.098 <sup>B</sup>	0.111 <sup>B</sup>	0.118 <sup>B</sup>	0.291 <sup>A</sup>	0.096 <sup>B</sup>	0.098 <sup>B</sup>	0.116 <sup>B</sup>	0.115 <sup>B</sup>	0.120 <sup>B</sup>	0.123 <sup>B</sup>	0.0105	***
c9 C12:1	0.868 <sup>B</sup>	0.812 <sup>CD</sup>	0.812 <sup>CDE</sup>	0.759 <sup>E</sup>	0.796 <sup>CDE</sup>	0.787 <sup>CDE</sup>	0.697 <sup>F</sup>	0.773 <sup>DE</sup>	0.995 <sup>A</sup>	0.881 <sup>B</sup>	0.876 <sup>B</sup>	0.827 <sup>BC</sup>	0.0210	***
C13:0	0.976 <sup>ABC</sup>	0.927 <sup>CD</sup>	0.935 <sup>DE</sup>	0.752 <sup>F</sup>	0.745 <sup>F</sup>	0.768 <sup>F</sup>	0.713 <sup>F</sup>	0.824 <sup>E</sup>	1.026 <sup>A</sup>	0.970 <sup>ABCD</sup>	1.001 <sup>AB</sup>	0.933 <sup>BCD</sup>	0.0316	***
C14:0 iso	0.837 <sup>DE</sup>	0.818 <sup>E</sup>	0.846 <sup>CDE</sup>	1.008 <sup>A</sup>	1.048 <sup>A</sup>	1.004 <sup>A</sup>	0.907 <sup>B</sup>	0.882 <sup>BCD</sup>	0.913 <sup>BC</sup>	0.864 <sup>CDE</sup>	0.863 <sup>CDE</sup>	0.891 <sup>BCDE</sup>	0.0416	***
C14:0	116 <sup>B</sup>	113 <sup>C</sup>	109 <sup>DE</sup>	107 <sup>E</sup>	109 <sup>D</sup>	109 <sup>DE</sup>	105 <sup>F</sup>	110 <sup>D</sup>	123 <sup>A</sup>	117 <sup>B</sup>	117 <sup>B</sup>	116 <sup>B</sup>	1.1	***
t9 C14:1	2.23 <sup>DEF</sup>	2.23 <sup>DE</sup>	2.40 <sup>BC</sup>	2.65 <sup>A</sup>	2.74 <sup>A</sup>	2.63 <sup>A</sup>	2.41 <sup>B</sup>	2.31 <sup>CD</sup>	2.27 <sup>DE</sup>	2.10 <sup>F</sup>	2.17 <sup>EF</sup>	2.21 <sup>DEF</sup>	0.072	***
C15:0 anteiso	4.26 <sup>BCD</sup>	4.36 <sup>BCD</sup>	5.07 <sup>A</sup>	5.14 <sup>A</sup>	5.23 <sup>A</sup>	4.50 <sup>B</sup>	4.52 <sup>BC</sup>	4.48 <sup>BC</sup>	4.34 <sup>BCD</sup>	4.00 <sup>D</sup>	4.15 <sup>CD</sup>	4.18 <sup>BCD</sup>	0.120	***
c9 C14:1	9.29 <sup>DE</sup>	8.64 <sup>F</sup>	8.17 <sup>G</sup>	8.66 <sup>F</sup>	9.18 <sup>DE</sup>	9.08 <sup>E</sup>	8.99 <sup>E</sup>	9.61 <sup>B</sup>	10.99 <sup>A</sup>	9.73 <sup>B</sup>	9.57 <sup>BC</sup>	9.32 <sup>CD</sup>	0.136	***
C15:0	10.8 <sup>CD</sup>	10.1 <sup>E</sup>	10.0 <sup>EF</sup>	9.8 <sup>F</sup>	10.1 <sup>EF</sup>	10.1 <sup>E</sup>	10.6 <sup>E</sup>	11.0 <sup>BC</sup>	11.9 <sup>A</sup>	11.4 <sup>B</sup>	11.4 <sup>B</sup>	11.1 <sup>B</sup>	0.17	***
C16:0 iso	2.00 <sup>EF</sup>	2.02 <sup>EF</sup>	2.04 <sup>DE</sup>	2.26 <sup>AB</sup>	2.32 <sup>A</sup>	2.23 <sup>B</sup>	2.11 <sup>E</sup>	2.10 <sup>CD</sup>	2.02 <sup>EF</sup>	1.98 <sup>FG</sup>	1.96 <sup>G</sup>	2.06 <sup>EF</sup>	0.056	***
C16:0	341 <sup>A</sup>	321 <sup>C</sup>	289 <sup>E</sup>	297 <sup>D</sup>	299 <sup>D</sup>	305 <sup>D</sup>	319 <sup>C</sup>	329 <sup>B</sup>	346 <sup>A</sup>	348 <sup>A</sup>	344 <sup>A</sup>	342 <sup>A</sup>	3.6	***
t6+t7+t8 C16:1	0.309 <sup>CDE</sup>	0.303 <sup>DE</sup>	0.318 <sup>BCD</sup>	0.322 <sup>BC</sup>	0.333 <sup>AB</sup>	0.339 <sup>A</sup>	0.337 <sup>A</sup>	0.341 <sup>A</sup>	0.300 <sup>E</sup>	0.233 <sup>F</sup>	0.239 <sup>F</sup>	0.245 <sup>F</sup>	0.0068	***
t9 C16:1	0.077 <sup>C</sup>	0.089 <sup>BC</sup>	0.105 <sup>A</sup>	0.094 <sup>AB</sup>	0.086 <sup>BC</sup>	0.098 <sup>AB</sup>	0.081 <sup>C</sup>	0.077 <sup>CD</sup>	0.096 <sup>AB</sup>	0.063 <sup>D</sup>	0.077 <sup>C</sup>	0.089 <sup>BC</sup>	0.0050	***
C17:0 iso	3.50 <sup>F</sup>	3.76 <sup>E</sup>	4.78 <sup>AB</sup>	4.82 <sup>AB</sup>	4.91 <sup>A</sup>	4.65 <sup>BC</sup>	4.49 <sup>C</sup>	4.18 <sup>D</sup>	3.52 <sup>F</sup>	3.34 <sup>F</sup>	3.37 <sup>F</sup>	3.46 <sup>F</sup>	0.095	***
t11+t12+t13 C16:1	1.72 <sup>F</sup>	1.87 <sup>E</sup>	2.32 <sup>A</sup>	2.18 <sup>BC</sup>	2.19 <sup>BC</sup>	2.18 <sup>B</sup>	2.09 <sup>C</sup>	1.97 <sup>D</sup>	1.72 <sup>F</sup>	1.56 <sup>G</sup>	1.58 <sup>G</sup>	1.61 <sup>G</sup>	0.037	***
c9 C16:1 + C17 anteiso	18.3 <sup>CDE</sup>	17.9 <sup>EF</sup>	18.2 <sup>DC</sup>	18.4 <sup>DC</sup>	18.6 <sup>C</sup>	18.2 <sup>CDEF</sup>	19.1 <sup>B</sup>	19.3 <sup>A</sup>	19.3 <sup>AB</sup>	18.1 <sup>DEF</sup>	17.8 <sup>FG</sup>	17.7 <sup>G</sup>	0.31	***
c11 C16:1	1.66 <sup>BC</sup>	1.71 <sup>AB</sup>	1.66 <sup>BC</sup>	1.42 <sup>EF</sup>	1.54 <sup>DE</sup>	1.58 <sup>CD</sup>	1.82 <sup>A</sup>	1.70 <sup>BC</sup>	1.56 <sup>CD</sup>	1.25 <sup>F</sup>	1.32 <sup>F</sup>	1.28 <sup>F</sup>	0.097	***



c13 C16:1	1.45 <sup>BC</sup>	1.32 <sup>BC</sup>	1.30 <sup>BC</sup>	1.15 <sup>C</sup>	1.24 <sup>BC</sup>	1.17 <sup>C</sup>	1.22 <sup>BC</sup>	1.39 <sup>BC</sup>	1.87 <sup>A</sup>	1.50 <sup>B</sup>	1.47 <sup>B</sup>	1.40 <sup>BC</sup>	0.069	***
C17:0	5.21 <sup>AB</sup>	5.17 <sup>AB</sup>	5.38 <sup>AB</sup>	5.56 <sup>A</sup>	5.48 <sup>AB</sup>	5.36 <sup>AB</sup>	5.41 <sup>AB</sup>	5.34 <sup>AB</sup>	4.57 <sup>C</sup>	5.07 <sup>BC</sup>	5.07 <sup>BC</sup>	5.10 <sup>AB</sup>	0.160	***
t10 c17:1	0.529 <sup>E</sup>	0.546 <sup>DE</sup>	0.589 <sup>A</sup>	0.590 <sup>AB</sup>	0.582 <sup>ABC</sup>	0.561 <sup>BCD</sup>	0.566 <sup>BCD</sup>	0.561 <sup>CD</sup>	0.499 <sup>F</sup>	0.409 <sup>G</sup>	0.368 <sup>H</sup>	0.395 <sup>GH</sup>	0.0126	***
C18:0 iso	0.582 <sup>EFG</sup>	0.609 <sup>CDE</sup>	0.726 <sup>A</sup>	0.711 <sup>AB</sup>	0.753 <sup>A</sup>	0.658 <sup>BC</sup>	0.646 <sup>BCD</sup>	0.608 <sup>DEF</sup>	0.560 <sup>FG</sup>	0.549 <sup>FG</sup>	0.543 <sup>G</sup>	0.547 <sup>G</sup>	0.0325	***
c9 C17:1	2.15 <sup>D</sup>	2.15 <sup>D</sup>	2.38 <sup>A</sup>	2.37 <sup>A</sup>	2.38 <sup>A</sup>	2.27 <sup>BC</sup>	2.35 <sup>AB</sup>	2.25 <sup>C</sup>	2.11 <sup>D</sup>	1.94 <sup>E</sup>	1.92 <sup>E</sup>	1.92 <sup>E</sup>	0.041	***
C18:0	97 <sup>D</sup>	104 <sup>C</sup>	108 <sup>B</sup>	110 <sup>AB</sup>	109 <sup>B</sup>	108 <sup>B</sup>	111 <sup>A</sup>	104 <sup>C</sup>	90 <sup>E</sup>	97 <sup>D</sup>	97 <sup>D</sup>	99 <sup>D</sup>	1.8	***
t4 C18:1	0.129 <sup>DEF</sup>	0.130 <sup>DEF</sup>	0.141 <sup>BCD</sup>	0.125 <sup>DEF</sup>	0.110 <sup>EF</sup>	0.134 <sup>CDE</sup>	0.135 <sup>CDE</sup>	0.149 <sup>BCD</sup>	0.105 <sup>F</sup>	0.166 <sup>AB</sup>	0.157 <sup>ABC</sup>	0.179 <sup>A</sup>	0.0099	***
t5 C18:1	0.102 <sup>CDE</sup>	0.110 <sup>BCD</sup>	0.116 <sup>AB</sup>	0.112 <sup>ABC</sup>	0.095 <sup>E</sup>	0.100 <sup>DE</sup>	0.064 <sup>G</sup>	0.062 <sup>G</sup>	0.080 <sup>F</sup>	0.117 <sup>AB</sup>	0.095 <sup>E</sup>	0.122 <sup>A</sup>	0.0055	***
t6+t7+t8 C18:1	2.42 <sup>D</sup>	2.65 <sup>BC</sup>	2.83 <sup>A</sup>	2.76 <sup>AB</sup>	2.48 <sup>CD</sup>	2.65 <sup>B</sup>	2.86 <sup>A</sup>	2.92 <sup>A</sup>	2.46 <sup>CD</sup>	2.33 <sup>D</sup>	2.37 <sup>D</sup>	2.50 <sup>CD</sup>	0.083	***
t9 C18:1	1.60 <sup>E</sup>	1.82 <sup>CDE</sup>	2.01 <sup>A</sup>	1.87 <sup>ABC</sup>	1.80 <sup>BCD</sup>	1.83 <sup>BCD</sup>	1.44 <sup>F</sup>	1.37 <sup>F</sup>	1.68 <sup>DE</sup>	1.94 <sup>ABC</sup>	1.96 <sup>AB</sup>	2.04 <sup>A</sup>	0.074	***
t10 C18:1	2.94 <sup>E</sup>	4.07 <sup>BC</sup>	4.10 <sup>B</sup>	6.03 <sup>A</sup>	5.29 <sup>A</sup>	3.88 <sup>BCD</sup>	1.97 <sup>F</sup>	1.66 <sup>F</sup>	3.00 <sup>E</sup>	3.17 <sup>DE</sup>	3.31 <sup>CDE</sup>	3.25 <sup>CDE</sup>	0.288	***
t11 C18:1	10.1 <sup>E</sup>	12.2 <sup>D</sup>	20.9 <sup>A</sup>	15.9 <sup>C</sup>	16.1 <sup>C</sup>	17.2 <sup>BC</sup>	18.6 <sup>B</sup>	17.7 <sup>BC</sup>	10.9 <sup>DE</sup>	9.9 <sup>E</sup>	10.2 <sup>E</sup>	10.5 <sup>DE</sup>	1.00	***
c6 + t12 C18:1	2.91 <sup>B</sup>	3.02 <sup>B</sup>	2.96 <sup>B</sup>	3.05 <sup>B</sup>	2.42 <sup>C</sup>	2.46 <sup>C</sup>	1.49 <sup>E</sup>	1.23 <sup>E</sup>	1.83 <sup>D</sup>	2.91 <sup>B</sup>	3.32 <sup>A</sup>	3.00 <sup>AB</sup>	0.134	***
c9 C18:1	186 <sup>D</sup>	195 <sup>C</sup>	209 <sup>B</sup>	212 <sup>B</sup>	211 <sup>B</sup>	211 <sup>B</sup>	223 <sup>A</sup>	213 <sup>B</sup>	183 <sup>DE</sup>	178 <sup>E</sup>	179 <sup>E</sup>	184 <sup>DE</sup>	2.3	***
t15 C18:1	1.84 <sup>B</sup>	1.87 <sup>B</sup>	2.07 <sup>B</sup>	2.07 <sup>B</sup>	1.82 <sup>B</sup>	1.98 <sup>B</sup>	0.95 <sup>C</sup>	0.58 <sup>D</sup>	1.27 <sup>C</sup>	3.07 <sup>A</sup>	3.13 <sup>A</sup>	3.12 <sup>A</sup>	0.111	***
c11 C18:1	4.82 <sup>FG</sup>	5.36 <sup>BCD</sup>	5.75 <sup>A</sup>	5.43 <sup>B</sup>	5.08 <sup>CDEF</sup>	5.14 <sup>CDEF</sup>	5.38 <sup>BC</sup>	5.41 <sup>B</sup>	4.58 <sup>G</sup>	4.95 <sup>EF</sup>	5.03 <sup>DEF</sup>	5.21 <sup>BCDE</sup>	0.251	***
c12 C18:1	2.23 <sup>AB</sup>	2.27 <sup>AB</sup>	1.91 <sup>C</sup>	1.93 <sup>C</sup>	1.66 <sup>D</sup>	1.87 <sup>C</sup>	1.88 <sup>C</sup>	1.95 <sup>C</sup>	1.93 <sup>C</sup>	2.26 <sup>B</sup>	2.30 <sup>AB</sup>	2.43 <sup>A</sup>	0.100	***
c13 C18:1	0.809 <sup>EF</sup>	0.902 <sup>BCD</sup>	1.019 <sup>A</sup>	0.886 <sup>BC</sup>	0.824 <sup>DE</sup>	0.897 <sup>BC</sup>	0.939 <sup>B</sup>	0.901 <sup>B</sup>	0.750 <sup>F</sup>	0.833 <sup>CDE</sup>	0.785 <sup>EF</sup>	0.837 <sup>CDE</sup>	0.0250	***
t16 + c14 C18:1	3.16 <sup>DE</sup>	3.51 <sup>B</sup>	3.72 <sup>A</sup>	3.44 <sup>BC</sup>	3.33 <sup>CD</sup>	3.39 <sup>BC</sup>	3.52 <sup>B</sup>	3.28 <sup>D</sup>	2.85 <sup>G</sup>	2.93 <sup>FG</sup>	3.04 <sup>EF</sup>	3.06 <sup>EF</sup>	0.049	***
c15 C18:1 + C19:0	1.43 <sup>B</sup>	1.50 <sup>AB</sup>	1.59 <sup>AB</sup>	1.59 <sup>AB</sup>	1.58 <sup>AB</sup>	1.41 <sup>B</sup>	1.63 <sup>A</sup>	1.55 <sup>AB</sup>	1.18 <sup>C</sup>	0.75 <sup>D</sup>	0.78 <sup>D</sup>	0.77 <sup>D</sup>	0.042	***
t11t15 C18:2	0.215 <sup>EF</sup>	0.330 <sup>D</sup>	0.583 <sup>A</sup>	0.435 <sup>BC</sup>	0.479 <sup>BC</sup>	0.474 <sup>BC</sup>	0.493 <sup>AB</sup>	0.418 <sup>C</sup>	0.242 <sup>DE</sup>	0.155 <sup>EF</sup>	0.199 <sup>EF</sup>	0.150 <sup>F</sup>	0.0387	***
t9t12 C18:2	0.042 <sup>E</sup>	0.073 <sup>BCDE</sup>	0.110 <sup>AB</sup>	0.057 <sup>DE</sup>	0.064 <sup>BCDE</sup>	0.085 <sup>ABCD</sup>	0.114 <sup>A</sup>	0.098 <sup>ABC</sup>	0.048 <sup>DE</sup>	0.042 <sup>DE</sup>	0.060 <sup>CDE</sup>	0.035 <sup>E</sup>	0.0124	***
c9t13 C18:2	1.84 <sup>D</sup>	2.07 <sup>C</sup>	2.58 <sup>A</sup>	2.17 <sup>BC</sup>	2.16 <sup>BC</sup>	2.22 <sup>B</sup>	2.30 <sup>B</sup>	2.22 <sup>B</sup>	1.85 <sup>D</sup>	1.61 <sup>E</sup>	1.68 <sup>DE</sup>	1.74 <sup>DE</sup>	0.052	***
c10t14 C18:2	1.13 <sup>AB</sup>	1.18 <sup>A</sup>	1.14 <sup>ABC</sup>	1.07 <sup>BCD</sup>	1.03 <sup>BCD</sup>	1.04 <sup>D</sup>	1.04 <sup>D</sup>	1.12 <sup>AB</sup>	1.04 <sup>BCD</sup>	1.01 <sup>D</sup>	1.03 <sup>D</sup>	1.03 <sup>CD</sup>	0.040	***
c9t14 C18:2	1.13 <sup>E</sup>	1.22 <sup>D</sup>	1.31 <sup>AB</sup>	1.25 <sup>BCD</sup>	1.23 <sup>CD</sup>	1.27 <sup>BC</sup>	1.35 <sup>A</sup>	1.31 <sup>AB</sup>	1.14 <sup>E</sup>	0.98 <sup>G</sup>	1.04 <sup>F</sup>	1.04 <sup>FG</sup>	0.026	***
c9t12 C18:2	0.579 <sup>DEF</sup>	0.656 <sup>AB</sup>	0.679 <sup>A</sup>	0.629 <sup>BC</sup>	0.590 <sup>DE</sup>	0.610 <sup>CD</sup>	0.669 <sup>A</sup>	0.639 <sup>BC</sup>	0.555 <sup>F</sup>	0.551 <sup>F</sup>	0.569 <sup>EF</sup>	0.586 <sup>DEF</sup>	0.0121	***
c16 C18:1	0.256 <sup>D</sup>	0.352 <sup>C</sup>	0.500 <sup>A</sup>	0.427 <sup>B</sup>	0.404 <sup>B</sup>	0.393 <sup>B</sup>	0.424 <sup>B</sup>	0.413 <sup>B</sup>	0.312 <sup>C</sup>	0.209 <sup>E</sup>	0.239 <sup>DE</sup>	0.232 <sup>DE</sup>	0.0173	***
t11c15 C18:2	1.22 <sup>E</sup>	1.56 <sup>D</sup>	2.64 <sup>A</sup>	2.29 <sup>BC</sup>	2.44 <sup>B</sup>	2.31 <sup>B</sup>	2.25 <sup>BC</sup>	2.09 <sup>C</sup>	1.44 <sup>DE</sup>	1.26 <sup>E</sup>	1.28 <sup>E</sup>	1.22 <sup>E</sup>	0.143	***
t9c12 C18:2	0.224 <sup>A</sup>	0.250 <sup>A</sup>	0.140 <sup>BC</sup>	0.154 <sup>B</sup>	0.109 <sup>CD</sup>	0.113 <sup>CD</sup>	0.170 <sup>B</sup>	0.225 <sup>A</sup>	0.239 <sup>A</sup>	0.080 <sup>D</sup>	0.095 <sup>D</sup>	0.073 <sup>D</sup>	0.0180	***
c9c12 C18:2	17.4 <sup>AB</sup>	18.1 <sup>A</sup>	17.2 <sup>ABC</sup>	16.8 <sup>BCD</sup>	15.5 <sup>EF</sup>	16.4 <sup>CDE</sup>	16.9 <sup>BC</sup>	17.2 <sup>ABC</sup>	15.7 <sup>EF</sup>	15.6 <sup>F</sup>	15.5 <sup>F</sup>	16.1 <sup>DEF</sup>	0.56	***
t12c15 C18:2 + c9 C19:1	0.237 <sup>G</sup>	0.290 <sup>DEF</sup>	0.350 <sup>C</sup>	0.339 <sup>CD</sup>	0.330 <sup>CDE</sup>	0.275 <sup>EFG</sup>	0.319 <sup>CDE</sup>	0.312 <sup>CDE</sup>	0.253 <sup>FG</sup>	0.894 <sup>B</sup>	0.938 <sup>AB</sup>	0.986 <sup>A</sup>	0.0220	***
C20:0	1.54 <sup>B</sup>	1.44 <sup>CD</sup>	1.33 <sup>E</sup>	1.51 <sup>BC</sup>	1.42 <sup>D</sup>	1.30 <sup>E</sup>	1.48 <sup>BCD</sup>	1.48 <sup>CD</sup>	1.32 <sup>E</sup>	1.62 <sup>A</sup>	1.66 <sup>A</sup>	1.68 <sup>A</sup>	0.043	***
c6c9c12 C18:3	0.221 <sup>BC</sup>	0.239 <sup>ABC</sup>	0.306 <sup>A</sup>	0.244 <sup>ABC</sup>	0.230 <sup>BC</sup>	0.223 <sup>BC</sup>	0.227 <sup>BC</sup>	0.231 <sup>BC</sup>	0.196 <sup>C</sup>	0.274 <sup>AB</sup>	0.239 <sup>ABC</sup>	0.250 <sup>ABC</sup>	0.0120	*
c8 C20:1	1.03 <sup>CD</sup>	1.00 <sup>D</sup>	0.93 <sup>E</sup>	1.08 <sup>BC</sup>	1.09 <sup>B</sup>	1.07 <sup>BC</sup>	1.14 <sup>A</sup>	1.16 <sup>A</sup>	1.04 <sup>BCD</sup>	1.03 <sup>BCD</sup>	1.07 <sup>BC</sup>	1.10 <sup>B</sup>	0.026	***
c11 C20:1	0.428 <sup>D</sup>	0.471 <sup>BC</sup>	0.465 <sup>BC</sup>	0.478 <sup>AB</sup>	0.438 <sup>CD</sup>	0.446 <sup>CD</sup>	0.497 <sup>AB</sup>	0.516 <sup>A</sup>	0.442 <sup>CD</sup>	0.079 <sup>E</sup>	0.075 <sup>E</sup>	0.046 <sup>E</sup>	0.0147	***
c9c12c15 C18:3	4.69 <sup>D</sup>	5.20 <sup>C</sup>	6.54 <sup>A</sup>	6.09 <sup>B</sup>	6.07 <sup>B</sup>	5.99 <sup>B</sup>	6.11 <sup>B</sup>	5.51 <sup>C</sup>	4.60 <sup>D</sup>	4.52 <sup>D</sup>	4.63 <sup>D</sup>	4.72 <sup>D</sup>	0.380	***

c9t11 C18:2	4.76 <sup>E</sup>	5.65 <sup>C</sup>	8.83 <sup>A</sup>	8.31 <sup>AB</sup>	8.52 <sup>AB</sup>	8.44 <sup>AB</sup>	8.36 <sup>AB</sup>	7.97 <sup>B</sup>	5.62 <sup>CD</sup>	4.76 <sup>E</sup>	4.92 <sup>DE</sup>	4.93 <sup>DE</sup>	0.409	***
Unknown C18:2 conjugated	0.289 <sup>C</sup>	0.282 <sup>C</sup>	0.356 <sup>B</sup>	0.484 <sup>A</sup>	0.383 <sup>B</sup>	0.352 <sup>B</sup>	0.385 <sup>B</sup>	0.352 <sup>B</sup>	0.294 <sup>C</sup>	0.197 <sup>D</sup>	0.167 <sup>DE</sup>	0.146 <sup>E</sup>	0.0185	***
Unknown C18:2 conjugated	0.223 <sup>F</sup>	0.249 <sup>EF</sup>	0.372 <sup>CBA</sup>	0.351 <sup>BCD</sup>	0.392 <sup>AB</sup>	0.384 <sup>A</sup>	0.385 <sup>AB</sup>	0.382 <sup>ABC</sup>	0.268 <sup>EF</sup>	0.275 <sup>E</sup>	0.324 <sup>D</sup>	0.344 <sup>CD</sup>	0.0283	***
c11c14 C20:2	0.103 <sup>F</sup>	0.116 <sup>EF</sup>	0.125 <sup>E</sup>	0.125 <sup>EF</sup>	0.126 <sup>EF</sup>	0.170 <sup>D</sup>	0.227 <sup>C</sup>	0.222 <sup>C</sup>	0.188 <sup>D</sup>	0.349 <sup>B</sup>	0.338 <sup>B</sup>	0.375 <sup>A</sup>	0.0091	***
C22:0	0.617 <sup>DE</sup>	0.622 <sup>CD</sup>	0.623 <sup>CD</sup>	0.678 <sup>AB</sup>	0.583 <sup>EF</sup>	0.536 <sup>G</sup>	0.709 <sup>A</sup>	0.712 <sup>A</sup>	0.569 <sup>FG</sup>	0.665 <sup>BC</sup>	0.618 <sup>DE</sup>	0.652 <sup>BCD</sup>	0.0320	***
c8c11c14 C20:3	0.764 <sup>AB</sup>	0.768 <sup>AB</sup>	0.753 <sup>AB</sup>	0.745 <sup>ABC</sup>	0.699 <sup>CD</sup>	0.729 <sup>BC</sup>	0.749 <sup>AB</sup>	0.772 <sup>A</sup>	0.685 <sup>D</sup>	0.764 <sup>AB</sup>	0.764 <sup>AB</sup>	0.782 <sup>A</sup>	0.0265	***
c13 C22:1	0.115	0.131	0.149	0.136	0.123	0.125	0.128	0.221	0.175	0.167	0.246	0.150	0.0266	†
c11c14c17 C20:3	0.078 <sup>E</sup>	0.089 <sup>CDE</sup>	0.111 <sup>CD</sup>	0.118 <sup>C</sup>	0.116 <sup>CD</sup>	0.109 <sup>CDE</sup>	0.117 <sup>C</sup>	0.101 <sup>CDE</sup>	0.083 <sup>DE</sup>	0.189 <sup>B</sup>	0.221 <sup>B</sup>	0.307 <sup>A</sup>	0.0155	***
c5c8c11c14 C20:4	0.98 <sup>BC</sup>	0.99 <sup>BC</sup>	0.97 <sup>CD</sup>	0.96 <sup>CD</sup>	0.91 <sup>D</sup>	0.95 <sup>CD</sup>	1.00 <sup>BC</sup>	1.03 <sup>AB</sup>	0.91 <sup>D</sup>	1.09 <sup>A</sup>	1.10 <sup>A</sup>	1.05 <sup>AB</sup>	0.033	***
c13c16 C22:2	0.364 <sup>FG</sup>	0.398 <sup>EF</sup>	0.521 <sup>A</sup>	0.470 <sup>BCD</sup>	0.493 <sup>AB</sup>	0.426 <sup>DE</sup>	0.458 <sup>BC</sup>	0.435 <sup>CDE</sup>	0.340 <sup>G</sup>	0.433 <sup>CDE</sup>	0.467 <sup>BC</sup>	0.459 <sup>BC</sup>	0.0233	***
c5c8c11c14c17 C20:5	0.502 <sup>CD</sup>	0.513 <sup>CD</sup>	0.636 <sup>A</sup>	0.641 <sup>AB</sup>	0.659 <sup>A</sup>	0.644 <sup>AB</sup>	0.647 <sup>A</sup>	0.611 <sup>B</sup>	0.535 <sup>C</sup>	0.515 <sup>CD</sup>	0.488 <sup>D</sup>	0.484 <sup>D</sup>	0.0327	***
C24:0	0.360 <sup>E</sup>	0.366 <sup>DE</sup>	0.377 <sup>CDE</sup>	0.428 <sup>A</sup>	0.411 <sup>AB</sup>	0.425 <sup>A</sup>	0.423 <sup>A</sup>	0.410 <sup>AB</sup>	0.333 <sup>F</sup>	0.409 <sup>AB</sup>	0.388 <sup>CD</sup>	0.394 <sup>BC</sup>	0.0192	***
c13c16c19 C22:3	0.071 <sup>E</sup>	0.078 <sup>DE</sup>	0.080 <sup>DE</sup>	0.091 <sup>CDE</sup>	0.079 <sup>DE</sup>	0.109 <sup>C</sup>	0.095 <sup>CDE</sup>	0.096 <sup>CD</sup>	0.075 <sup>DE</sup>	0.184 <sup>A</sup>	0.154 <sup>B</sup>	0.163 <sup>AB</sup>	0.0071	***
c7c10c13c16 C22:4	0.129 <sup>CDE</sup>	0.148 <sup>CD</sup>	0.124 <sup>DE</sup>	0.118 <sup>DEF</sup>	0.102 <sup>EEFG</sup>	0.181 <sup>BC</sup>	0.081 <sup>FG</sup>	0.071 <sup>G</sup>	0.090 <sup>FG</sup>	0.231 <sup>A</sup>	0.242 <sup>A</sup>	0.205 <sup>AB</sup>	0.0155	***
c7c10c13c16c19 C22:5	0.786 <sup>D</sup>	0.787 <sup>D</sup>	0.878 <sup>BC</sup>	0.895 <sup>BC</sup>	0.934 <sup>AB</sup>	0.919 <sup>ABC</sup>	0.980 <sup>A</sup>	0.960 <sup>AB</sup>	0.937 <sup>AB</sup>	0.856 <sup>CD</sup>	0.918 <sup>BC</sup>	0.881 <sup>BC</sup>	0.0441	***
c4c7c10c13c16c19 C22:6	0.059	0.058	0.067	0.066	0.066	0.059	0.132	0.079	0.061	0.070	0.063	0.058	0.0087	ns

<sup>a</sup> In September, there was a missing sample of free-range milk in the analysis of basic composition and the mean on this set of parameters was calculated from nine samples

<sup>b</sup> Significances were declared at \*\*\*, P < 0.001; \*\*, P < 0.01; \*, P < 0.05; †, 0.05 < P < 0.10 (trend); ns, P > 0.10 (non-significant). Means for month within a row with different upper case letters are significantly different according to Fisher's Least Significant Difference test (P < 0.05)