Effect of short-term versus long-term grassland management and seasonal variation in organic and conventional dairy farming on the composition of bulk tank milk

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ABSTRACT

Bulk tank milk from 28 dairy farms was sampled every second month for 2 yr to assess the effects of grassland management, production system and season on milk fatty acid (FA) composition, concentrations of fat-soluble vitamins, Se, and milk sensory quality. Grassland management varied in terms of time since establishment. Short-term grassland management (SG) was defined as establishment or reseeding every fourth year or more often, and long-term grassland management (LG) was defined as less frequent establishment or reseeding. Fourteen organic (ORG) dairy farms with either short-term or long-term grassland management were paired with 14 conventional (CON) farms with respect to grassland management. Within ORG farms, SG farms differed from LG farms in herbage botanical composition, but not in concentrate FA concentrations, dry matter intake, or milk yield. Within CON farms, herbage composition, concentrate FA concentrations, dry matter intake, and milk yield showed no or insignificant variations. The ORG farms differed from CON farms in herbage botanical composition, concentrate FA concentrations, concentrate intake, and milk yield. Compared with ORG-LG farms, ORG-SG farms produced milk fat with higher proportions of C10:0 and C12:0 associated with higher herbage proportions of legumes (*Fabaceae*) and lower proportions of other dicotyledon families. Compared with milk from CON farms, milk fat from ORG farms had higher proportions of most saturated FA and all n-3 FA, but lower proportions of C18:0 and C18:1 *cis*-9 associated with higher forage proportion and differences in concentrations of FA in concentrates. Compared with the outdoor-feeding periods, the indoor feeding periods yielded milk fat with higher proportions of most short-chain and medium-chain FA and lower proportions of most C18-FA associated with grazing and higher forage proportions. Milk concentrations of α-tocopherol and β-carotene were lower during the grazing periods. Inclusion of fishmeal in organic concentrates may explain higher Se concentrations in organically produced milk. Milk sensory quality was not affected in this study. In conclusion, grassland management had minor effects on milk composition, and differences between ORG farms and CON farms may be explained by differences in concentrate intake and concentrate FA concentrations. Milk produced on ORG farms versus CON farms and milk produced during the outdoor versus indoor feeding periods had potential health benefits due to FA composition. In contrast, the higher milk-fat proportions of saturated FA in milk from ORG farms may be perceived as negative for human health.
Key words: dairy farm, grassland management, production system, milk composition

INTRODUCTION

Consumption of dairy products has been claimed to have negative health effects in humans because milk fat has high proportions of SFA, which are understood to contribute to cardiovascular disease and obesity (Appleby et al., 1999; Insel et al., 2007). In contrast, new studies have revealed that high consumption of dairy products may help to prevent coronary heart disease, different types of cancer, and other chronic diseases, although the mechanisms are not understood (Kliem and Givens, 2011). Fat-soluble vitamins are important in human nutrition (Haug et al., 2007) and, besides their nutritional value, they may also improve the oxidative stability of milk fat with high proportions of PUFA (Al-Mabruk et al., 2004). Diet formulation may influence the milk-fat composition of grazing or silage-fed cows (Chilliard et al., 2001).

Grassland management is likely to affect sward botanical composition, which has been proven to affect milk FA composition (Dewhurst et al., 2003b; Lourenço et al., 2008). Sward botanical composition affects the

concentrations of fat-soluble vitamins in forages, but also other factors, such as plant maturity (Danielsson et al., 2008) and preservation method (Lindqvist et al., 2012), are important. Milk concentrations of fat-soluble vitamins vary significantly and depend on diet, animal factors, such as breed (McDowell, 2000) and stage of lactation (Jensen et al., 1999), and supplementation. Forage Se concentrations have been reported inadequate to meet the dietary Se requirement in regions with low soil Se concentrations, and thus Se-enriched feed supplements are commonly used (Govasmark et al., 2005).

Compared with conventionally produced milk, organically produced milk has higher fat proportions of C18:3n-3 (Butler et al., 2008; Collomb et al., 2008a), C18:1 *trans*-11 and C18:2 *cis*-9,*trans*-11 (Jahreis et al., 1996; Collomb et al., 2008a), PUFA (Ellis et al., 2006; Collomb et al., 2008a), and lower proportions of n-6 FA (Butler et al., 2008; Collomb et al., 2008a;), C18:0, C18:1 *cis*-9, and MUFA (Jahreis et al., 1996; Collomb et al., 2008a). The concentrations of α-tocopherol and β-carotene are higher in organic than conventional milk during the outdoor feeding period, but not during the indoor feeding period in the study of Butler et al. (2008). Ellis et al. (2007) reported no differences for these vitamins, but found less retinol in organic milk. The concentrations of α-tocopherol and β-carotene are higher in summer milk than in winter milk (Lindmark-Månsson et al., 2003; Ellis et al., 2007). In these studies, the authors suggest a positive effect of grazing compared with feeding silage on fat-soluble vitamins in milk, however, the role of forage type and total diet needs further investigations.

Sward age and composition on dairy farms vary according to conditions that complicate soil tillage necessary for reseeding, such as climate, slope, stone content, field size, or economic costs. The time span after reseeding and other grassland management factors may affect grassland botanical composition (Hopkins, 1986), which, together with differences in dietary supplementation, affect milk FA composition (Dewhurst et al., 2003b), concentrations of fat-soluble vitamins (Bolstad et al., 2007), and Se concentrations in milk. Although previously cited studies indicate specific effects of botanical composition on milk FA and fat-soluble vitamins, to our knowledge no attempts have been made to investigate the effect of botanical composition at the level of farming systems (Ellis et al., 2007; Butler et al., 2008; Collomb et al., 2008a).

The objectives of the present study were to compare the effects of long-term versus short-term grassland management in organic and conventional production systems, compare organic and conventional production systems, and assess seasonal variation on FA composi-

tion, fat-soluble vitamin concentrations, sensory quality, and Se concentration in bulk tank milk.

MATERIALS AND METHODS

Experimental Design

Twenty-eight dairy farms in central Norway participated in the study in 2007 and 2008. Seven organic (**ORG**) farms with short-term grassland management (**SG**), referred to as ORG-SG farms, were paired with 7 conventional (**CON**) farms with SG, referred to as CON-SG farms, and 7 ORG farms with long-term grassland management (**LG**), referred to as ORG-LG farms, were paired with 7 CON farms with LG, referred to as CON-LG farms. Grassland management was defined as SG when the grassland fields of a farm were renewed every fourth year or more frequently and as LG when the fields were renewed less frequently. Fields were renewed by soil tillage and seeding. Organic and conventional farms were paired on location and calving pattern, based on information from local extension services. The organic farms were certified by the Norwegian certification body Debio (Bjørkelangen, Norway) according to the EU standards for organic farming (European Commission, 2006). In brief, the standards for organic farming require a minimum forage intake in total DMI (50% in the first 3 mo of lactation increasing to 60% thereafter) and all feeds have to be grown organically (i.e., without the use of synthetic pesticides and synthetic N fertilizers). Fertilization with animal manure is limited to 170 kg of N/ha and year. All farms participated in the Norwegian Dairy Herd Recording System and delivered milk to the same dairy company (TINE Norwegian Dairies SA, Oslo, Norway). On all farms, forages were fed ad libitum and allocated concentrate amounts were based on individual milk yields.

On-Farm Analysis, Sampling, and Data Collection

Data on farm characteristics (Table 1) were collected in farmer interviews and milk production data were collected from the Norwegian Dairy Herd Recording System. Herbage botanical composition before first cut silage in 2007 was estimated on 4 selected fields on each farm by the dry-weight-rank method (Mannetje and Haydock, 1963), modified by Jones and Hargreaves (1979). The selected fields represented overall grassland use including fields that were cut, cut and grazed in combination, or only grazed. Silage and concentrates were sampled twice in each indoor feeding period (February 2007, October 2007, February 2008, December 2008) on each farm. Two milk samples were taken in each sampling month (February, April, June, August,

Table 1. Characteristics of dairy farms with an organic production system and short-term (ORG-SG) or long-term grassland management (ORG-LG) and dairy farms with a conventional production system and short-term (CON-SG) or long-term grassland management (CON-LG; n = 7)

Item	Farming system							
	ORG-SG		ORG-LG		CON-SG		CON-LG	
	Mean	SD	Mean	SD	Mean	SD	Mean	SD
Average distance between paired farms,[1] km	3.0	2.04	6.5	7.09				
Proportion of calvings[1,2]								
January to March	0.26	0.192	0.21	0.139	0.15	0.092	0.19	0.141
April to June	0.26	0.254	0.18	0.120	0.34	0.242	0.24	0.283
July to September	0.26	0.139	0.35	0.172	0.29	0.172	0.27	0.133
October to December	0.23	0.125	0.27	0.084	0.22	0.111	0.30	0.225
Farm area, ha	32	6.6	32	11.1	27	12.1	22	4.8
Altitude, m	69	39.5	106	71.6	53	38.9	141	125.1
Herd size, cows	22	9.5	15	3.0	18	4.8	19	6.3
Forage area proportion	0.87	0.055	0.99	0.023	0.85	0.213	1.00	0.000
Grassland age,[3] yr	3	1.0	11	3.6	3	0.9	10	4.1
Date of first cut silage	12 June	5.5	22 June	10.8	13 June	4.8	18 June	7.9
Manure,[4] kg of total N/ha and yr	80	27.5	101	49.5	107	50.4	178	94.8
Fertilizer, kg of N/ha and yr	0		0		235	44.1	242	13.3

[1]Organic farms were paired with CON farms with respect to grassland management on location and calving pattern.
[2]From 2003 through 2008.
[3]Weighted for field area.
[4]Calculated as 3.1 kg of N/t of manure (Daugstad et al., 2012).

October, December) from stirred bulk tanks after 4 to 6 milkings and transported at 4°C to the dairy.

Animals and Diets

On most farms cows were housed in tiestalls, and on 6 farms, mainly SG farms, in loose-house barns. The cows were mainly fed grass silage-based diets during the indoor feeding periods (October to mid-May), and on all farms cows grazed in the outdoor feeding periods, although many herds also had access to silage. Only herds with Norwegian Red dairy cows were included in the study, and calving time was rather evenly distributed over the year on all farms. Dairy feed rations included forage, concentrates, mineral mixtures, and vitamin mixtures on all farms. Silage fermented in bulk silos or in round bales (proportion of silage fermented in round bales in total silage produced: ORG-SG = 0.36, ORG-LG = 0.20, CON-SG = 0.58, CON-LG = 0.29) was the main forage in the indoor feeding periods. On most farms the herbage was wilted before ensiling. The cows were kept indoors at night on 8 farms (ORG-SG: 1, ORG-LG: 3, CON-SG: 1, CON-LG: 3) during the outdoor feeding periods. Homegrown grains of barley and oats were fed in addition to commercial concentrates on most ORG-SG farms and supplemented with fishmeal on 2 ORG-SG farms.

Commercial concentrates were used on all farms, but ingredients varied; organic mixes contained (on average) barley 29%, wheat 25%, oats 21%, fishmeal 7%, sugarcane molasses 5%, and expeller soybean meal 4%, and conventional mixes contained (on average) barley 36%, oats 15%, solvent-extracted soybean meal 12%, Sorghum (*Sorghum bicolor* (L.) Moench) 10%, rape seeds and expeller rape seed meal 8%, sugar beet pulp 8%, sugarcane molasses 7%, rumen protected fat (saturated vegetable FA, mainly C16:0; AkoFeed Gigant 60; Aarhus Karlshamn AB, Malmö, Sweden) 2%, and vegetable fat (vegetable FA, mainly C18:1 *cis*-9, C16:0; C18:2n-6; AkoFeed Standard; Aarhus Karlshamn AB) 1%. Additionally, on some farms, other feed supplements, such as potato (1 ORG-SG farm), whey (1 ORG-LG farm, 1 CON-SG farm), brewers' grain (1 CON-SG farm), or macroalgae meal (1 ORG-SG farm), were used.

Commercial concentrates contained synthetic vitamins (vitamin A: 1.2–1.5 mg/kg; vitamin E: 30–50 mg/kg) and mineral sodiumselenite (25–40 mg Se/kg). Additionally, most farms supplemented the dairy cows with about 100 g/cow of mineral mixtures (e.g., Natura Minovit Drøv and Pluss Multitilskudd, Felleskjøpet Agri, Gardermoen, Norway), containing synthetic vitamins [vitamin A (0.12 mg/g), vitamin E (2.0 mg/g)] and mineral sodiumselenite (25 μg of Se/g). Fishmeal used in organic commercial concentrates or given as supplement contained 3.6 mg of Se/kg of DM, although no information is available on the daily amount of mineral mixtures or fishmeal fed.

Analysis Methods

Sample Preparation and Chemical Analysis of Forage and Concentrates. Feed samples were stored

at −20°C, freeze-dried (Christ LCM-2, Beta 1–16 and Christ LOC-1m, Alpha 1–4, Martin Christ, Osterode am Harz, Germany; Hetosicc, Birkerød, Denmark), milled (1.0-mm screen; Retsch SM 100, Retsch GmbH, Haan, Germany), and then re-stored at −20°C in plastic bags before analysis of chemical constituents. Forage was analyzed at the Dairy One Inc. Forage Testing Laboratory (Ithaca, NY) for ash by the AOAC method (942.05.), N by the AOAC method (990.03), soluble protein by sodium borate-sodium phosphate buffer procedure on a TruMac N Macro Determinator (Leco, St. Joseph, MI; Roe and Sniffen, 1990), ether extract by the AOAC method (2003.05; AOAC, 1990), NDF by the method of Van Soest et al. (1991) with heat-stable amylase and sodium sulfite, and in vitro true digestibility by the ANKOM DaisyII Filter Bag Technique (Ankom Technology, Macedon, NY) after incubation for 48 h.

Feed FA were extracted by the Röse-Gottlieb method (Molkentin and Precht, 1995) and analyzed as FA methyl esters (**FAME**) on GC (Thermo Finnigan Focus, Thermo Fisher Scientific Inc., Waltham, MA), equipped with a column of 105 m × 0.25 mm i.d. and 0.2 μm of film thickness (Restek RTX-2330, Restek Corporation, Bellefonte, PA). Fatty acids were quantified by comparison with external standards.

Sample Preparation and Chemical Analysis of Milk. Milk samples intended for analysis of FA composition, fat-soluble vitamin concentrations, and Se concentrations were frozen (−20°C) immediately after arriving at the dairy. Milk samples intended for analysis of gross composition, urea, and FFA were preserved with 2-bromo-2-nitropropane-1,3-diol (Bronopol, D&F Inc., Dublin, CA) and analyzed by a Fourier transformed infrared spectroscopy milk analyzer (MilkoScan 6000 FTIR, Foss, Hillerød, Denmark). Milk samples intended for assessment of sensory quality were stored at 4°C. For analysis of FA, fat was extracted from milk with chloroform and methanol according to Bligh and Dyer (1959) and analyzed as FAME, as described by Jensen and Nielsen (1996) by GC (Hewlett Packard 6890 series, Agilent Technologies, Palo Alto, CA) equipped with an automatic on-column injector (Hewlett Packard 7673; split ratio 4.325:1), a capillary column of 30 m × 320 μm i.d. and 0.25-μm film thickness (Omegawax, Supelco 4–293–415, Sigma-Aldrich, St. Louis, MO), and a flame-ionization detector. The FA C17:0 was used as external standard. Fat-soluble vitamins were extracted and analyzed by HPLC as described by Jensen and Nielsen (1996) and Jensen et al. (1998). A PerkinElmer HS-5-Silica column (4.0 × 125 mm; Waltham, MA) was used for analyses of α-tocopherol and retinol and a Supelco amino column (4.6 × 250 mm; Sigma-Aldrich) was used for analysis of β-carotene. Total milk Se con-

centrations were determined by inductively coupled plasma mass spectrometry (PerkinElmer, Elan 6000). Five milliliters of fresh milk were digested at 240°C for 40 min using an ultraclave (UltraCLAVE 3, Milestone, Shelton, CT) in 3.5 mL of distilled ultrapure HNO_3 (Merck, Whitehouse Station, NJ). The samples were dried and redissolved in 20 mL of 2% ethanol and 1% HNO_3 and stored for 6 wk before analysis. Tellur was used as internal standard. The standard reference material whole milk powder 8435 (0.131 SD, 0.014 μg Se/g) from the National Institute of Standards and Technology (Gaithersburg, MD) was digested with HNO_3 in triplicate and analyzed for quality assurance and control purposes. The relative standard deviation based on counting statistics was less than 2%.

Assessment of Sensory Quality. Test panels of at least 2 trained assessors (TINE Norwegian Dairies SA) evaluated raw bulk tank milk odor and taste on a 5-point scale, where 1 = milk with serious deviation from normal taste and 5 = milk with no deviation from normal taste. Milk samples were assessed the day after delivery to the dairy plants.

Calculations

Estimates of feed intake were based on the participating herds' data in the Norwegian Dairy Herd Recording System. Daily concentrate DMI per cow was weighted by milk production on farm level as (sum of individual concentrate intakes × sum of individual milk yields)/(sum of individual milk yields). Daily forage net energy intake per cow was estimated as net energy requirement for maintenance (0.0424 × 600 kg of $BW^{0.75}$; (Van Es, 1978), added net energy requirement for milk production (0.44 × ECM + 0.0007293 × ECM^2; Van Der Honing and Alderman, 1988), and subtracted net energy intake of concentrates (individual concentrate intakes × net energy concentration based on product declaration). Forage DMI was estimated by dividing the estimated forage net energy intake by the energy concentration in forage samples, estimated from in vitro true digestibility analyses. In the indoor feeding periods for months without feed samples, average values were calculated, and in the outdoor feeding periods a feed table value was used (NE_L: 6.86 SD, 0.386 MJ/ kg of DM; Anonymous, 2012). Apparent de novo FA synthesis was calculated as the daily secretion of the sum of C4:0 to C14:1c9 plus the sum of C15:0 to 17:1c9 × 0.5, as a share (approximately 50%) of the latter FA is blood derived.

Statistical Analysis

Feed composition, DMI, milk yield, milk composition, and milk sensory quality were analyzed using the

MIXED model procedure by SAS (SAS, 2009). Statistical model 1 was

$$Y_{ijklmn} = \mu + G_i + P(G)_{ij} + M_k + (GM)_{ik} + [MP(G)]_{ijk} \\ + f(G,P)_{ijl} + t(G)_{im} + e_{ijklmn},\qquad [1]$$

where Y were the individual dependent variables for DMI, feed composition, milk yield, and milk composition ($n = 1$–336); μ was the average of all observations; G was the fixed effect of grassland management ($i = 1$, 2; where $1 = $ SG and $2 = $ LG); $P(G)$ was the fixed effect of production system within G ($j = 1$, 2; where $1 = $ ORG and $2 = $ CON); M was the fixed effect of month ($k = 1$–12; where $1 = $ February 2007, $2 = $ April 2007, $3 = $ June 2007, $4 = $ August 2007, $5 = $ October 2007, $6 = $ December 2007, $7 = $ February 2008, $8 = $ April 2008, $9 = $ June 2008, $10 = $ August 2008, $11 = $ October 2008, $12 = $ December 2008); (GM) and $[MP(G)]$ were interactions of the fixed effects; f was the random effect of farm within G and P ($l = 1$ through 28); t was the random effect of farm pair within G ($m = 1$–14); and e_{ijklmn} were the random residual errors, assumed to be independent and $N\left(0, \sigma_e^2\right)$. Observations for month within farm were treated as repeated observations. Contrasts were calculated for the effects of ORG-SG versus ORG-LG, CON-SG versus CON-LG, ORG versus CON, and indoor ($k = 1, 2, 5, 6, 7, 8, 11, 12$) versus outdoor feeding periods ($k = 3, 4, 9, 10$). Differences between means were tested with the Tukey-Kramer test. For analysis of feed chemical composition the statistical model 2 was

$$Y_{ijklmn} = \mu + G_i + P(G)_{ij} + m_k + f(G,P)_{ijl} \\ + t(G)_{im} + s(m,f)_{ijkl} + e_{ijklmn},\qquad [2]$$

where Y, μ, G, $P(G)$, $f(G,P)$, $t(G)$, and e were as specified for model 1; m was the random effect of month ($k = 1, 5, 7, 12$; number of month as specified for M in model 1); and s was the random effect of sample within m and f ($l = 1$–113). For analysis of botanical composition the GLIMMIX procedure in SAS (SAS, 2009), which fits generalized linear mixed models, was used. Statistical model 2 was applied with the modification that $s(f)$ was the number of fields within farm assessed ($l = 1$–4). To find correlations between proportions of botanical families in the herbage, concentrate DMI and milk-fat proportions of selected FA a principal component analysis was performed by the PRINCOMP procedure in SAS (SAS, 2009).

RESULTS

Botanical and Chemical Composition of Diets

The herbage botanical composition of fields that were cut or both cut and grazed differed between ORG-SG and ORG-LG farms and between ORG farms and CON farms, although differences between CON-SG and CON-LG farms were small (Table 2). Grassland on ORG-SG farms contained more ($P = 0.001$) legumes and less ($P = 0.009$) dandelion than on ORG-LG farms, and grassland on ORG farms contained less ($P = <0.001$) grass and more ($P = <0.001$) legumes than on CON farms. For fields solely used as pastures, no differences were found between farming systems. In pastures, the proportions averaged 655 g of DM/kg of DM for grasses, 53 g of DM/kg of DM for legumes, and 292 g of DM/kg of DM for other dicotyledons.

Despite differences in the botanical composition of fields used for silage production, small differences were found in silage chemical composition between farming systems (Table 3). Compared with silages from ORG-LG, silages from ORG-SG had lower ($P = 0.04$) concentrations of C18:3n-3. Compared with silages from CON, silages from ORG had less CP ($P < 0.001$) and crude fat ($P = 0.04$) and more NFC ($P < 0.001$). The FA composition of concentrates did not differ between ORG-SG and ORG-LG or between CON-SG and CON-LG, but large differences were found between ORG and CON (Table 4). Compared with concentrates used on CON farms, concentrates used on ORG farms had less ($P < 0.001$) C18:0 and C18:1 cis-9, and more ($P < 0.001$) C18:3n-3 and C22:6n-3.

Effects of Grassland Management and Production System on Feed Intake, Milk Yield, and Milk Composition

Grassland management had small effects on DMI, daily milk yields, and milk composition, whereas production system had pronounced effects. Compared with milk from ORG-LG, milk from ORG-SG had higher protein concentrations ($P < 0.05$) and higher milk-fat proportions of the SFA C9:0, C10:0, C11:0, C12:0 ($P \leq 0.01$), C14:1 cis-9 ($P = 0.005$), C22:1 cis-11 ($P = 0.005$), and C22:6n-3 ($P < 0.001$), and lower proportions of C17:1 cis-9 ($P = 0.01$), C18:4n-3 ($P = 0.007$) and C24:0 ($P = 0.005$), whereas only small differences were found in milk from CON-SG compared with CON-LG (Tables 5 and 6). The secretion of the apparently de novo synthesized FA C10:0 through C14:1 cis-9 was higher ($P = 0.04$) for ORG-SG than for ORG-LG (Table 7). Grass-

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Table 2. Herbage botanical composition of fields for cutting and combined cutting or grazing (g of DM/kg of DM) before first cut in 2007 on dairy farms with an organic production system (ORG) and short-term (ORG-SG) or long-term grassland management (ORG-LG) and dairy farms with conventional production system (CON) and short-term (CON-SG) or long-term grassland management (CON-LG; 7 farms × 3 fields; n = 21)

| Item | Farming system | | | | | P-value[1] | | | | |
	ORG-SG	ORG-LG	CON-SG	CON-LG	SEM	G[2]	P(G)[3]	ORG-SG vs. ORG-LG[4]	CON-SG vs. CON-LG[5]	ORG vs. CON[6]
Grasses (*Poaceae*; 19 spp.)	642^{bc}	505^{c}	918^{a}	818^{ab}	53.5	0.037	<0.001	0.084	NS	<0.001
Timothy (*Phleum pratense* L.)	342^{ab}	128^{b}	495^{a}	249^{b}	51.6	<0.001	0.061	0.007	0.003	0.021
Meadow fescue (*Festuca pratensis* Huds.)	101	79	154	141	41.8	NS	NS	NS	NS	NS
Perennial ryegrass (*Lolium perenne* L.)	69	31	97	22	29.5	0.067	NS	NS	0.087	NS
Smooth meadowgrass (*Poa pratensis* L.)	50	44	40	48	31.0	NS	NS	NS	NS	NS
Rough meadowgrass (*Poa trivialis* L.)	25	45	29	126	27.2	0.078	0.093	NS	0.020	0.099
Common couch (*Elytrigia repens* L.)	34	8	64	112	27.9	NS	0.037	NS	NS	0.024
Other grasses	20	169	37	121	35.9	0.013	NS	0.007	NS	NS
Legumes (*Fabaceae*; 8 spp.)	265^{a}	141^{b}	32^{c}	15^{c}	23.3	0.006	<0.001	0.001	NS	<0.001
Red clover (*Trifolium pratense* L.)	174^{a}	94^{ab}	20^{b}	12^{b}	23.4	0.062	<0.001	0.018	NS	<0.001
White clover (*Trifolium repens* L.)	58^{a}	49^{ab}	12^{ab}	3^{b}	12.5	NS	0.011	NS	NS	0.003
Other legumes	33	0	0	0	9.3	0.095	0.069	0.022	NS	0.091
Other botanical families (20 spp.)	94^{b}	357^{a}	51^{b}	167^{ab}	56.7	0.003	0.072	0.003	NS	0.051
Northern dock (*Rumex longifolius* DC.)	36	94	15	40	24.6	NS	NS	NS	NS	NS
Dandelion (*Taraxacum* spp.)	33^{ab}	92^{a}	14^{b}	27^{b}	14.5	0.034	0.016	0.009	NS	0.010
Common sorrel (*Rumex acetosa* L.)	3	71	1	56	28.4	0.090	NS	NS	NS	NS
Creeping buttercup (*Ranunculus repens* L.)	8	48	2	22	14.2	0.048	NS	0.060	NS	NS
Meadow buttercup (*Ranunculus acris* L.)	5	22	2	4	7.3	NS	NS	NS	NS	NS
Other spp.	7	32	17	18	11.0	NS	NS	NS	NS	NS
Number of spp. per field	15.1^{ab}	17.3^{a}	12.3^{b}	13.2^{b}	0.87	0.092	0.002	0.091	NS	0.001

$^{a-c}$Means within a row with different letters differ (Tukey-Kramer test, $P < 0.05$).

[1]Nonsignificant = $P > 0.10$.

[2]Effect of grassland management.

[3]Effect of production system within grassland management.

[4]Contrast of ORG-SG vs. ORG-LG.

[5]Contrast of CON-SG vs. CON-LG.

[6]Contrast of ORG vs. CON.

Table 3. Silage chemical composition, feed values, and FA composition on dairy farms with an organic production system (ORG) and short-term (ORG-SG) or long-term grassland management (ORG-LG) and dairy farms with a conventional production system (CON) and short-term (CON-SG) or long-term grassland management (CON-LG)

| Item | Farming system | | | | | P-value[1] | | | | |
	ORG-SG	ORG-LG	CON-SG	CON-LG	SEM	G[2]	P(G)[3]	ORG-SG vs. ORG-LG[4]	CON-SG vs. CON-LG[5]	ORG vs. CON[6]
n	31	25	27	29						
Chemical composition and feed value										
DM, g/kg	285	271	258	254	19.8	NS	NS	NS	NS	NS
CP, g/kg of DM	136[b]	140[b]	170[a]	168[a]	6.7	NS	<0.001	NS	NS	<0.001
Soluble protein, g/kg of CP	606[ab]	532[b]	638[a]	595[ab]	23.7	0.058	0.049	0.036	NS	0.021
Crude fat, g/kg of DM	44	51	52	55	3.8	NS	0.085	0.085	NS	0.038
NDF, g/kg of DM	555	561	574	589	14.5	NS	NS	NS	NS	0.075
ADF, g/kg of DM	366	374	373	384	8.4	NS	NS	NS	NS	NS
NFC, g/kg of DM	223[a]	214[a]	174[b]	176[b]	13.8	NS	0.001	NS	NS	<0.001
OM, g/kg of DM	922[ab]	927[ab]	918[b]	936[a]	5.4	0.065	NS	NS	0.012	NS
In vitro true digestibility (48 h), g/kg of DM	822	802	820	808	10.9	NS	NS	NS	NS	NS
NE$_L$, MJ/kg of DM	5.7	5.6	5.7	5.7	0.11	NS	NS	NS	NS	NS
FA, g/kg of DM										
C16:0	3.86	4.21	4.13	4.19	0.605	NS	NS	NS	NS	NS
C18:0	0.39	0.41	0.35	0.36	0.062	NS	NS	NS	NS	0.076
C18:1 *cis*-9	0.79	1.10	0.69	0.84	0.177	0.057	NS	0.047	NS	0.089
C18:2n-6	3.66	4.29	3.75	3.82	0.660	NS	NS	0.083	NS	NS
C18:3n-3	8.25	10.25	10.71	10.35	1.603	NS	0.042	0.044	NS	0.067
Total FA	22.8	26.9	26.5	26.8	3.67	NS	NS	0.048	NS	NS

[a–c]Means within a row with different letters differ (Tukey-Kramer test, $P < 0.05$).

[1]Nonsignificant = $P > 0.10$.

[2]Effect of grassland management.

[3]Effect of production system within grassland management.

[4]Contrast of ORG-SG vs. ORG-LG.

[5]Contrast of CON-SG vs. CON-LG.

[6]Contrast of ORG vs. CON.

Table 4. Concentrate FA composition on dairy farms with an organic production system (ORG) and short-term (ORG-SG) or long-term grassland management (ORG-LG) and dairy farms with a conventional production system (CON) and short-term (CON-SG) or long-term grassland management (CON-LG) during the indoor feeding periods

	Farming system					P-value[1]				
Item	ORG-SG	ORG-LG	CON-SG	CON-LG	SEM	G[2]	P(G)[3]	ORG-SG vs. ORG-LG[4]	CON-SG vs. CON-LG[5]	ORG vs. CON[6]
n	29	27	29	27						
FA, g/kg DM										
C16:0	7.66	7.56	10.75	10.67	1.096	NS	0.025	NS	NS	0.008
C18:0	0.55[c]	0.62[bc]	0.93[ab]	1.18[a]	0.094	NS	<0.001	NS	0.072	<0.001
C18:1 *cis*-9	9.55[b]	10.06[b]	17.29[a]	18.66[a]	1.826	NS	<0.001	NS	NS	<0.001
C18:2n-6	15.31	14.76	17.78	15.52	2.134	NS	NS	NS	NS	NS
C18:3n-3	0.77[a]	0.60[a]	0.26[b]	0.29[b]	0.080	NS	<0.001	0.077	NS	<0.001
C20:5n-3	0.38[a]	0.31[a]	0.01[b]	0.03[b]	0.038	NS	<0.001	NS	NS	<0.001
C22:6n-3	0.60[a]	0.49[a]	0.02[b]	0.05[b]	0.059	NS	<0.001	NS	NS	<0.001
Total FA	39.79[bc]	39.21[c]	52.75[a]	51.36[ab]	4.401	NS	0.004	NS	NS	0.001

[a–c]Means within a row with different letters differ (Tukey-Kramer test, $P < 0.05$).

[1]Nonsignificant = $P > 0.10$.

[2]Effect of grassland management.

[3]Effect of production system within grassland management.

[4]Contrast of ORG-SG vs. ORG-LG.

[5]Contrast of CON-SG vs. CON-LG.

[6]Contrast of ORG vs. CON.

land management within the production system did not affect milk concentrations of fat-soluble vitamins, Se, and milk sensory quality (Table 8).

Compared with cows on CON farms, cows on ORG farms had lower daily concentrate intakes (means = ORG: 4.5 kg of DM/d, CON: 6.4 kg of DM/d; $P = 0.001$), lower milk yields (ORG: 20.0 kg/d; CON: 22.5 kg/d; $P = 0.03$), and lower milk urea concentrations (ORG: 4.0 mmol/L, CON: 5.4 mmol/L; $P < 0.001$). Milk fat from ORG farms had higher proportions of most SFA, C18:1 *trans* (includes several coeluting trans FA) FA (ORG: 29.2 g/kg of FAME, CON: 25.3 g/kg of FAME; $P = 0.03$) and all n-3 FA than milk fat from CON-farms. This resulted in a lower n-6:n-3 FA ratio in milk from ORG farms (2.1 vs. 3.0; $P < 0.001$). The proportions of C18:0 (ORG: 103.0 g/kg of FAME, CON: 129.8 g/kg of FAME; $P < 0.001$) and C18:1 *cis*-9 (ORG: 214.7 g/kg of FAME, CON: 2.44.8 g/kg of FAME; $P < 0.001$) were lower in milk fat from ORG farms than CON farms. Calculated as proportions in total C18-FA in milk fat, ORG farms had higher proportions of most unsaturated C18-FA and lower proportions of C18:0 ($P < 0.001$) compared with CON farms (Table 9). Milk concentrations of Se were higher ($P = 0.009$) on ORG farms than on CON farms. The concentrations of fat-soluble vitamins and sensory quality were not affected by production system.

Effect of Season on Feed Intake, Milk Yield, and Milk Composition

Total DMI was higher ($P < 0.001$) during the indoor feeding season than the outdoor feeding season, but for-

age proportion and daily milk yields were not affected. Milk concentrations of fat ($P < 0.001$) and FFA ($P < 0.001$) were higher during the indoor feeding periods. Compared with milk from the outdoor feeding periods, milk from the indoor feeding periods had higher proportions of most short-chain and medium-chain SFA and lower proportions of most C18-FA, total MUFA, total PUFA, total n-6 FA, and total n-3 FA. The concentrations of α-tocopherol ($P < 0.001$) and β-carotene ($P = 0.02$) were higher during the indoor feeding periods than during outdoor feeding. Season did not affect milk Se concentrations and milk sensory quality.

The effects of grassland management and production system on milk FA composition were consistent across season, in general. For some FA, significant interaction effects were observed between month and management factors; however, these interactions did not alter the general effects of month or management factors.

Correlation of Herbage Botanical Composition and FA Composition in Milk

In the principal component analysis of herbage botanical composition, which examined concentrate DMI and the proportions of selected FA in milk fat, the principal component 1 explained 43% of the total variation and generally differentiated ORG farms from CON farms with the exception of 1 CON-LG farm (Figure 1). Principal component 2 explained 24% of the total variation and differentiated most ORG-SG farms from ORG-LG farms, but not CON-SG farms from CON-LG farms. The herbage proportion of the

Table 5. Average DMI, milk yield, and milk gross composition on dairy farms with an organic production system (ORG) and short-term (ORG-SG) or long-term grassland management (ORG-LG) and dairy farms with a conventional production system (CON) and short-term (CON-SG) or long-term grassland management (CON-LG) during indoor (IN) and outdoor feeding periods (OUT)

Item	ORG-SG	ORG-LG	CON-SG	CON-LG	SEM	IN	OUT	SEM	G[2]	P (G)[3]	M[4]	G × M[5]	P(G) × M[6]	ORG-SG vs. ORG-LG[7]	CON-SG vs. CON-LG[8]	ORG vs. CON[9]	IN vs. OUT[10]
											Farming system				Season						*P*-value[1]								
n	84	84	84	84		224	112										
Forage proportion,[11, 12] g/kg of total DMI	705[a]	699[a]	646[ab]	585[b]	24.6	658	660	20.6	NS	0.003	NS	NS	NS	NS	0.094	0.001	NS
DMI,[11] kg/d																	
Forage[12]	11.5	11.2	11.7	9.5	0.58	11.4	10.2	0.45	0.061	NS	<0.001	NS	NS	NS	0.012	NS	<0.001
Concentrates	4.6[b]	4.4[b]	6.2[ab]	6.5[a]	0.46	5.7	4.9	0.30	NS	0.004	<0.001	NS	NS	NS	NS	0.001	<0.001
Total	16.0[ab]	15.4[b]	17.8[a]	15.9[ab]	0.58	16.9	15.0	0.50	NS	0.004	<0.001	NS	NS	NS	0.024	0.057	<0.001
Milk yield, kg/d	20.9[ab]	19.0[b]	23.9[a]	21.1[ab]	1.13	21.1	21.5	0.82	0.048	0.095	NS	NS	NS	NS	0.094	0.034	NS
ECM,[13] kg/d	21.6[ab]	19.8[b]	24.8[a]	21.8[ab]	1.13	22.0	22.0	0.80	0.048	0.089	NS	NS	NS	NS	0.073	0.034	NS
Fat, g/kg	41.2	39.4	40.9	40.5	0.61	40.9	39.6	0.51	NS	NS	<0.001	NS	NS	0.053	NS	NS	<0.001
Protein, g/kg	34.7[a]	33.0[b]	34.2[ab]	33.4[ab]	0.36	33.8	33.8	0.33	0.021	NS	0.031	NS	NS	0.004	NS	NS	NS
Lactose, g/kg	47.0	46.6	46.7	46.8	0.20	46.6	47.1	0.16	NS	NS	<0.001	NS	NS	NS	NS	NS	<0.001
FFA, mEq/L	0.46	0.53	0.52	0.58	0.040	0.56	0.46	0.034	NS	NS	<0.001	NS	NS	NS	NS	NS	<0.001
Urea, mmol/L	4.2[b]	3.8[b]	5.4[a]	5.5[a]	0.21	4.7	4.7	0.17	NS	<0.001	<0.001	0.084	0.013	NS	NS	<0.001	NS

[a,b]Means within a row with different letters differ (Tukey-Kramer test, $P < 0.05$).

[1]Nonsignificant = $P > 0.10$.

[2]Effect of grassland management.

[3]Effect of production system within grassland management.

[4]Effect of month.

[5]Interaction between grassland management and month.

[6]Interaction between production system within grassland management and month.

[7]Contrast of ORG-SG vs. ORG-LG.

[8]Contrast of CON-SG vs. CON-LG.

[9]Contrast of ORG vs. CON.

[10]Contrast of IN vs. OUT.

[11]Weighted for individual milk yield.

[12]Estimates based on dairy cows' net energy requirements for maintenance and production.

[13]Calculated as kilograms of milk × (0.01 + 0.0122 × g of fat/kg of milk + 0.0077 × g of protein/kg of milk + 0.0053 × g of lactose/kg of milk).

Table 6. Fatty acid composition and total FA in milk from dairy farms with an organic production system (ORG) and short-term (ORG-SG) or long-term grassland management (ORG-LG) and dairy farms with a conventional production system (CON) and short-term (CON-SG) or long-term grassland management (CON-LG) during the indoor (IN) and outdoor feeding periods (OUT)

FA, g/kg of FA methyl esters	Farming system					Season			P-value[1]								
	ORG-SG	ORG-LG	CON-SG	CON-LG	SEM	IN	OUT	SEM	G[2]	P(G)[3]	M[4]	G × M[5]	P(G) × M[6]	ORG-SG vs. ORG-LG[7]	CON-SG vs. CON-LG[8]	ORG vs. CON[9]	IN vs. OUT[10]
n	84	84	84	84		224	112										
C4:0	38.3	39.1	37.7	40.1	0.75	39.1	38.1	1.07	0.072	NS	<0.001	NS	NS	NS	0.035	NS	0.096
C6:0	23.6	22.8	22.4	22.8	0.50	23.3	22.1	0.48	NS	NS	<0.001	NS	NS	NS	NS	NS	<0.001
C8:0	14.3	13.4	13.2	13.3	0.48	13.6	13.5	0.51	NS	0.086	<0.001	0.019	NS	NS	NS	0.078	NS
C9:0	0.1[a]	0.1[b]	0.1[b]	0.1[b]	0.01	0.1	0.1	0.01	0.067	0.013	<0.001	0.092	0.008	0.010	NS	0.021	<0.001
C10:0	33.0[a]	28.9[b]	28.9[b]	27.7[b]	0.94	30.0	28.9	0.86	0.034	0.005	<0.001	NS	NS	0.006	NS	0.003	0.009
C11:0	0.4[a]	0.3[b]	0.3[b]	0.2[b]	0.02	0.3	0.2	0.03	0.038	0.002	<0.001	NS	NS	0.008	NS	0.002	0.001
C12:0	37.6[a]	32.9[b]	32.2[b]	30.5[b]	1.12	33.8	32.2	0.98	0.032	0.001	<0.001	NS	0.054	0.007	NS	0.001	0.001
C13:0	1.0[a]	0.9[a]	0.8[a]	0.8[b]	0.03	0.9	0.8	0.04	NS	<0.001	<0.001	NS	NS	NS	NS	<0.001	0.001
C14:0	124.2[a]	117.4[ab]	112.4[bc]	108.8[c]	2.38	119.2	108.7	2.11	0.098	<0.001	<0.001	NS	NS	NS	0.057	<0.001	<0.001
C14:1 cis-9	10.6[a]	9.5[b]	9.3[b]	8.7[b]	0.25	10.0	8.6	0.29	0.002	<0.001	<0.001	NS	NS	0.005	0.088	<0.001	<0.001
C15:0	11.7[ab]	12.3[a]	10.2[c]	10.5[bc]	0.33	11.5	10.5	0.29	NS	<0.001	<0.001	NS	NS	NS	NS	<0.001	<0.001
C16:0	308.7[a]	305.0[a]	280.8[b]	277.8[b]	4.65	306.1	266.9	4.34	NS	<0.001	<0.001	NS	NS	NS	NS	<0.001	<0.001
C16:1 cis-9	15.8[a]	16.0[a]	14.0[b]	13.5[b]	0.45	14.7	15.1	0.41	NS	<0.001	0.004	NS	NS	NS	NS	<0.001	0.035
C16:1 cis-7	1.6	1.5	1.6	1.8	0.13	1.5	1.9	0.21	NS	NS	NS	NS	NS	NS	NS	NS	0.003
C17:1 cis-9	3.5[ab]	4.0[a]	3.3[b]	3.4[b]	0.13	3.5	3.7	0.11	0.0208	0.009	<0.001	NS	0.023	0.014	NS	0.004	<0.001
C18:0	99.8[b]	106.3[b]	127.6[a]	132.0[a]	3.17	112.7	123.9	2.68	0.0956	<0.001	<0.001	NS	NS	NS	NS	<0.001	<0.001
C18:1 cis-9	208.3[b]	221.2[b]	244.1[a]	245.5[a]	5.20	221.0	247.2	4.92	NS	<0.001	<0.001	NS	NS	0.096	NS	<0.001	<0.001
C18:1 cis-11	8.9	8.3	9.0	8.9	0.32	8.4	9.6	0.30	NS	NS	<0.001	NS	NS	NS	NS	NS	<0.001
C18:1 $trans$ FA	28.9	29.5	24.6	26.0	1.77	21.8	35.3	1.40	NS	0.100	<0.001	0.049	0.008	NS	NS	0.034	<0.001
C18:2n-6	18.1	17.8	19.2	18.4	0.60	17.5	20.1	0.45	NS	NS	<0.001	NS	NS	NS	NS	NS	<0.001
C18:2 cis-9,$trans$-11	6.9	7.6	6.8	6.4	0.57	6.0	8.7	0.44	NS	NS	<0.001	NS	NS	NS	NS	NS	<0.001
C18:2 $trans$-10,cis-12	0.2	0.3	0.2	0.2	0.05	0.2	0.2	0.05	NS	NS	<0.001	0.020	NS	0.062	NS	NS	0.024
C18:3n-6	1.1	1.2	1.0	1.2	0.07	1.1	1.2	0.10	NS	NS	<0.001	0.021	NS	NS	NS	NS	0.035
C18:3n-3	7.0[a]	7.1[a]	5.3[b]	6.1[ab]	0.34	5.9	7.4	0.28	NS	0.002	<0.001	NS	NS	NS	NS	0.001	<0.001
C18:4n-3	0.4[ab]	0.6[a]	0.4[b]	0.4[b]	0.06	0.4	0.5	0.06	0.095	0.009	<0.001	NS	NS	0.007	NS	0.012	NS
C20:0	2.7[a]	2.7[a]	2.0[b]	2.2[b]	0.11	2.5	2.3	0.03	NS	<0.001	0.002	NS	0.033	NS	NS	<0.001	0.001
C20:1 cis-9	3.1[a]	3.1[a]	2.2[b]	2.4[b]	0.16	2.7	2.7	0.16	NS	<0.001	<0.001	NS	NS	NS	NS	<0.001	NS
C20:2n-6	0.3	0.3	0.2	0.3	0.04	0.3	0.2	0.05	NS	NS	<0.001	NS	NS	NS	NS	NS	<0.001
C20:3n-6	0.5[b]	0.6[ab]	0.7[a]	0.6[ab]	0.05	0.5	0.7	0.08	NS	0.022	NS	NS	NS	NS	NS	0.016	0.009
C20:3n-3	0.2	0.2	0.1	0.2	0.02	0.2	0.1	0.02	NS	NS	<0.001	NS	NS	NS	NS	0.045	<0.001
C20:4n-6	0.9[b]	1.0[ab]	1.1[a]	1.1[ab]	0.05	1.0	1.1	0.07	NS	0.030	NS	NS	NS	NS	NS	0.057	NS
C20:5n-3	0.9[a]	0.9[a]	0.6[b]	0.7[b]	0.03	0.8	0.8	0.03	NS	<0.001	<0.001	NS	0.086	NS	NS	0.040	<0.001
C22:0	1.3[a]	1.4[a]	0.8[b]	0.9[b]	0.07	1.1	1.1	0.09	NS	<0.001	NS	0.100	0.071	NS	NS	<0.001	NS
C22:1 cis-11	1.0[a]	0.8[b]	0.2[c]	0.2[c]	0.06	0.6	0.5	0.06	NS	<0.001	<0.001	NS	0.072	0.005	NS	<0.001	0.001
C22:1 cis-13	0.2	0.2	0.1	0.1	0.03	0.2	0.2	0.05	NS	0.025	NS	NS	NS	NS	NS	0.007	NS
C22:5n-6	0.4[c]	0.4[bc]	0.5[ab]	0.6[a]	0.03	0.5	0.5	0.03	NS	<0.001	<0.001	0.070	NS	NS	NS	<0.001	0.041
C22:5n-3	1.0[a]	1.0[a]	0.8[b]	0.9[ab]	0.05	0.9	0.9	0.04	NS	<0.001	<0.001	NS	NS	NS	0.080	<0.001	0.027

Continued

Table 6 (Continued). Fatty acid composition and total FA in milk from dairy farms with an organic production system (ORG) and short-term (ORG-SG) or long-term grassland management (ORG-LG) and dairy farms with a conventional production system (CON) and short-term (CON-SG) or long-term grassland management (CON-LG) during the indoor (IN) and outdoor feeding periods (OUT)

FA, g/kg of FA methyl esters	Farming system					Season			P-value[1]								
	ORG-SG	ORG-LG	CON-SG	CON-LG	SEM	IN	OUT	SEM	G[2]	P(G)[3]	M[4]	G ×M[5]	P(G) ×M[6]	ORG-SG vs. ORG-LG[7]	CON-SG vs. CON-LG[8]	ORG vs. CON[9]	IN vs. OUT[10]
C22:6n-3	1.0[a]	0.7[b]	0.1[c]	0.2[c]	0.05	0.5	0.4	0.05	0.0447	<0.001	0.043	NS	NS	<0.001	NS	<0.001	NS
C24:0	0.5[b]	0.7[a]	0.5[b]	0.5[b]	0.04	0.5	0.6	0.07	0.006	0.009	NS	NS	NS	0.005	NS	0.004	NS
C24:1 cis-15	0.1	0.2	0.1	0.1	0.05	0.1	0.2	0.08	NS	NS	NS	NS	NS	NS	NS	NS	NS

[a–c]Means within a row with different letters differ (Tukey-Kramer test, $P < 0.05$).
[1]Nonsignificant = $P > 0.10$.
[2]Effect of grassland management.
[3]Effect of production system within grassland management.
[4]Effect of month.
[5]Interaction between grassland management and month.
[6]Interaction between production system within grassland management and month.
[7]Contrast of ORG-SG vs. ORG-LG.
[8]Contrast of CON-SG vs. CON-LG.
[9]Contrast of ORG vs. CON.
[10]Contrast of IN vs. OUT.

grass family was positively correlated with concentrate DMI (r = 0.64; $P < 0.001$) and negatively correlated with herbage proportion of the knotweed family (*Polygonaceae*; r = −0.72; $P < 0.001$), the buttercup family (*Ranunculaceae*; r = −0.72; $P < 0.001$) and the aster family (*Asteraceae*; r = −0.73; $P < 0.001$). Furthermore, the herbage grass family was negatively correlated with the milk-fat proportions of C18:1 *trans* FA (r = −0.57; $P = 0.002$), C18:2 *cis*-9,*trans*-11 (r = −0.53; $P < 0.004$), and C18:3n-3 (r = −0.83; $P < 0.001$). The forage proportion of the legume family was positively correlated with the milk FA proportions of C12:0 (r = 0.65; $P < 0.001$), C14:0 (r = 0.63; $P < 0.001$), C16:0 (r = 0.60; $P < 0.001$), and C22:6n-3 (r = 0.82; $P < 0.001$), and negatively correlated with the milk FA proportions of C18:0 (r = 0.71; $P < 0.001$) and C18:1 *cis*-9 (r = 0.71; $P < 0.001$; Figure 2). The CON-LG farm differing from the other CON-LG farms in the principal component analysis had less grasses (258 g/kg of DM), less legumes (3 g/kg of DM), more nonlegume dicotyledons (739 g/kg of DM), and lower concentrate level (4.6 kg of DM/d) per cow compared with the other CON-LG farms. Thus, this CON-LG farm had a botanical composition similar to that of ORG-LG farms. Milk fat proportions of SFA (649 g/kg of FAME) were lower and MUFA (306 g/kg of FAME) and PUFA (46 g/kg of FAME) were higher in milk from this farm than the average of CON-LG farms.

DISCUSSION

The observed differences in botanical composition between the farming systems may be due to differences in seed mixtures used, harvesting management, N-fertilization level, or herbicide use. On CON farms, the use of synthetic N-fertilizers most likely boosted plant growth from early spring, which may explain higher proportions of grass and CP concentrations in silage compared with ORG farms. Slightly higher NE_L concentrations in silage and higher total DMI, and thereby higher total NE_L intake on ORG-SG farms than on ORG-LG farms, may have stimulated microbial protein synthesis, explaining the higher protein concentrations in milk from ORG-SG than ORG-LG farms, in accordance with Coulon and Rémond (1991). The lower silage concentrations of C18:3n-3 for ORG-SG compared with ORG-LG may be related to red clover proportion, because concentrations of C18:3n-3 tend to be lower in fresh legumes than fresh grasses (Boufaïed et al., 2003). It has also been shown that the loss of unsaturated FA during silage fermentation may be higher for legumes than grasses (Knicky et al., 2012).

Table 7. Total FA concentration in milk, calculated sums and ratios of FA, and desaturase index in milk from dairy farms with an organic production system (ORG) and short-term (ORG-SG) or long-term grassland management (ORG-LG) and dairy farms with a conventional production system (CON) and short-term (CON-SG) or long-term grassland management (CON-LG) during the indoor (IN) and outdoor feeding periods (OUT)

Item	Farming system					Season			P-value[1]								
	ORG-SG	ORG-LG	CON-SG	CON-LG	SEM	IN	OUT	SEM	G[2]	P(G)[3]	M[4]	G × M[5]	P(G) × M[6]	ORG-SG vs. ORG-LG[7]	CON-SG vs. CON-LG[8]	ORG vs. CON[9]	IN vs. OUT[10]
n	84	84	84	84		224	112										
Total FA, g/kg of milk	33.1	31.8	32.9	33.0	0.56	33.2	31.6	0.72	NS	NS	<0.001	NS	NS	NS	NS	NS	<0.001
Calculated sums of FA, g/kg of FA methyl esters																	
Total SFA	697.1ᵃ	684.1ᵃᵇ	669.8ᵇ	668.3ᵇ	6.42	694.7	650.0	5.68	NS	0.004	<0.001	NS	NS	NS	NS	0.001	<0.001
Total MUFA	264.0ᵇ	276.2ᵃᵇ	293.1ᵃ	294.7ᵃ	5.90	269.4	307.3	5.28	NS	<0.001	<0.001	NS	NS	NS	NS	<0.001	<0.001
Total PUFA	38.9	39.7	37.1	37.0	1.12	35.9	42.7	0.93	NS	NS	<0.001	NS	NS	NS	NS	0.052	<0.001
Total n-6 FA	21.3	21.3	22.8	22.1	0.68	21.0	23.7	0.52	NS	NS	<0.001	0.012	NS	NS	NS	NS	<0.001
Total n-3 FA	10.6ᵃ	10.5ᵃ	7.3ᵇ	8.4ᵇ	0.42	8.7	10.1	0.34	NS	<0.001	<0.001	NS	NS	NS	0.087	<0.001	<0.001
Apparently de novo synthesized FA,[11] g/d																	
C4:0 to C9:0	53	46	58	53	3.5	54	50	2.81	NS	NS	<0.001	NS	NS	NS	NS	0.073	0.038
C10:0 to C14:1 *cis*-9	141	114	144	123	8.7	135	121	6.35	0.022	NS	<0.001	NS	NS	0.038	NS	NS	<0.001
C15:0 to C17:1 *cis*-9	117	102	121	107	6.5	117	101	4.79	0.050	NS	<0.001	NS	NS	NS	NS	NS	<0.001
Calculated ratios of FA																	
n-6:n-3 FA	2.1ᵇ	2.1ᵇ	3.2ᵃ	2.8ᵃ	0.16	2.6	2.5	0.15	NS	<0.001	0.008	NS	NS	NS	0.051	<0.001	NS
Desaturase index																	
C14[12]	0.078	0.075	0.076	0.073	0.0016	0.077	0.073	0.0016	0.073	NS	<0.001	NS	NS	NS	NS	NS	<0.001
C16[13]	0.049	0.050	0.048	0.047	0.0014	0.046	0.054	0.0013	NS	NS	<0.001	NS	0.093	NS	NS	0.055	<0.001

a,bMeans within a row with different letters differ (Tukey-Kramer test, $P < 0.05$).

[1]Nonsignificant = $P > 0.10$.

[2]Effect of grassland management.

[3]Effect of production system within grassland management.

[4]Effect of month.

[5]Interaction between grassland management and month.

[6]Interaction between production system within grassland management and month.

[7]Contrast of ORG-SG vs. ORG-LG.

[8]Contrast of CON-SG vs. CON-LG.

[9]Contrast of ORG vs. CON.

[10]Contrast of IN vs. OUT.

[11]Σ de novo synthesized FA and for C15 to C17: 0.5 × Σ de novo synthesized FA.

[12]C14:1 *cis*-9/(C14:1 *cis*-9 + C14:0).

[13]C16:1 *cis*-9 × (C16:1 *cis*-9 + C16:0).

Table 8. Milk concentrations of fat-soluble vitamins, Se, and milk sensory quality from dairy farms with an organic production system (ORG) and short-term (ORG-SG) or long-term grassland management (ORG-LG) and dairy farms with a conventional production system (CON) and short-term (CON-SG) or long-term grassland management (CON-LG) during the indoor (IN) and outdoor feeding period (OUT)

Item	Farming system					Season			P-value[1]								
	ORG-SG	ORG-LG	CON-SG	CON-LG	SEM	IN	OUT	SEM	G[2]	P(G)[3]	M[4]	G × M[5]	P(G) × M[6]	ORG-SG vs. ORG-LG[7]	CON-SG vs. CON-LG[8]	ORG vs. CON[9]	IN vs. OUT[10]
n	84	84	84	84		224	112										
Fat-soluble vitamins, mg/kg																	
α-Tocopherol	0.72	0.82	0.80	0.79	0.035	0.83	0.69	0.038	NS	0.084	<0.001	NS	NS	0.061	NS	NS	<0.001
β-Carotene	0.18	0.18	0.20	0.19	0.010	0.19	0.18	0.008	NS	NS	<0.001	0.045	NS	NS	NS	0.081	0.022
Retinol	0.46	0.45	0.44	0.44	0.017	0.44	0.46	0.019	NS	NS	<0.001	NS	NS	NS	NS	NS	0.077
Se, μg/kg	17.4^a	15.6^{ab}	14.8^{ab}	13.5^b	0.78	15.1	15.7	0.704	0.082	0.029	<0.001	NS	0.014	NS	NS	0.009	NS
Sensory quality[11]	4.88	4.89	4.93	4.91	0.037	4.89	4.93	0.057	NS	NS	0.015	NS	NS	NS	NS	NS	NS

^{a,b}Means within a row with different letters differ (Tukey-Kramer test, $P < 0.05$).

[1]Nonsignificant = $P > 0.10$.

[2]Effect of grassland management.

[3]Effect of production system within grassland management.

[4]Effect of month.

[5]Interaction between grassland management and month.

[6]Interaction between production system within grassland management and month.

[7]Contrast of ORG-SG vs. ORG-LG.

[8]Contrast of CON-SG vs. CON-LG.

[9]Contrast of ORG vs. CON.

[10]Contrast of IN vs. OUT.

[11]Scale of 1 to 5 points, where 1 = milk with serious deviation from normal taste and 5 = milk with no deviation from normal taste.

Intake of CP determines milk urea concentrations to a large extent (Broderick, 2003). Higher forage proportions and lower forage CP concentrations resulted in lower intakes of energy and CP and, thereby, lower milk yields and milk urea concentrations on ORG farms than on CON farms.

Effects on Milk FA Composition

Small differences in milk FA composition between ORG-SG and ORG-LG and between CON-SG and CON-LG are in accordance with Dewhurst et al. (2003b) and Steinshamn and Thuen (2008), who compared red and white clover grass silages. In contrast to the present study, however, silage containing red clover has, in most studies, increased milk fat proportions of C18:3n-3 compared with silages containing white clover or grass (Steinshamn and Thuen, 2008; Moorby et al., 2009). A possible explanation may be that the differences in clover species proportions were too small in the present study. Higher milk fat proportions of C10:0 and C12:0 for ORG-SG versus ORG-LG may be related to increased de novo synthesis, and higher milk fat proportions of C22:1 *cis*-11 and C22:6n-3 may be related to higher intake of fishmeal, in accordance with Donovan et al. (2000).

The increased milk fat proportions of total SFA for ORG is in accordance with the results from the indoor feeding period in the study of Butler et al. (2008), whereas Ellis et al. (2006) and Collomb et al. (2008a) found no differences between organic and conventional bulk tank milk. Increased de novo synthesis can be caused by increased concentrate level (Dewhurst et al., 2003b) or by low-fiber diets (Alzahal et al., 2009). However, in the present study, the daily secretion of apparently de novo synthesized FA was not affected by production system. Higher proportions of SFA may also be caused by increased mobilization of C16:0 from adipose tissue due to longer periods of feeding below energy balance in early lactation (Palmquist et al., 1993). As the plane of nutrition was lower for ORG farms than on CON farms, the cows on ORG farms were likely to be in lower energy status, which may explain the higher proportions of C16:0 in milk from ORG farms than in the milk from CON farms.

Lower milk fat proportions of C18:0 and C18:1 *cis*-9 in milk from ORG farms than CON farms are in accordance with Jahreis et al. (1996) and Collomb et al. (2008a), and is most likely related to differences in quantity and quality of fat sources used in the concentrates. A lower proportion of C18:2n-6 is in accordance with Jahreis et al. (1996). Higher milk fat proportions of C18:3n-3 and total n-3 FA for ORG than CON are in accordance with Ellis et al. (2006) and Collomb et al.
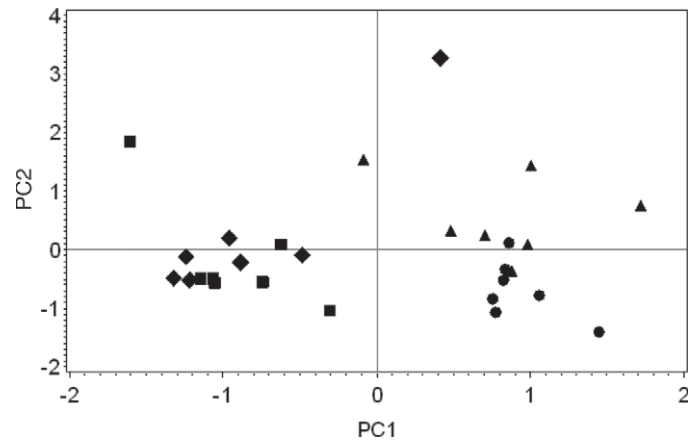
Figure 1. Score plot for first and second principal components (PC1 vs. PC2) for dairy farms with an organic production system and short-term (ORG-SG, ●) or long-term grassland management (ORG-LG, ▲) and dairy farms with a conventional production system and short-term (CON-SG, ■) or long-term grassland management (CON-LG, ◆) based on variables of milk fat proportions of selected FA, herbage proportions of botanical families, and cows' daily concentrate DMI (means over 2 yr for 28 farms).

(2008a), and may be explained by higher concentrate concentrations of C18:3n-3, higher dietary forage proportion (Dewhurst et al. 2003b; Steinshamn and Thuen 2008), or higher herbage proportions of red and white clover, which can reduce rumen biohydrogenation of C18:3n-3 as reported by Dewhurst et al. (2003a). Higher milk fat proportions of C22:5n-3 and C22:6n-3 may be explained by inclusion of fishmeal in concentrates used on ORG farms, in accordance with Donovan et al. (2000).

As forages did not differ in FA composition, and concentrates used on ORG farms contained less C18-FA than those used on CON farms, cows on ORG farms most likely had lower intakes of C18-FA. Therefore, to adjust for the difference in total C18-FA intake, the proportions of the different C18-FA in total C18-FA in milk were calculated. Lower proportions of C18:0 and higher proportions of C18:2n-6 and C18:3n-3 in total C18-FA in milk indicate that feed PUFA were to a lower extent biohydrogenated in the rumen of cows on ORG farms. Further, higher proportions of C18:1 *trans* FA and C18:2 *cis*-9,*trans*-11, partly deriving from the rumen biohydrogenation intermediate C18:1 *trans*-11, indicate an inhibition of the terminal biohydrogenation step in cows on ORG farms. An accumulation of biohydrogenation intermediates has been observed by including fish oil in the diet (Lee et al., 2008; Shingfield et al., 2012) or by feeding botanically diverse forages (Lourenço et al., 2008).

Seasonal effects on milk FA composition in this study confirm the findings of other studies (Elgersma

Table 9. Milk FA proportions in total C18-FA in milk from dairy farms with an organic (ORG) or a conventional production system (CON; n = 168)

FA, g/kg of total C18-FA methyl esters	Production system			P-value[1]
	ORG	CON	SEM	ORG vs. CON[2]
C18:0	278	305	5.2	<0.001
C18:1 *cis*-9	578	575	4.5	NS
C18:1 *cis*-11	23	21	0.7	0.004
C18:1 *trans* FA	78	59	3.7	<0.001
C18:2n-6	49	44	1.5	0.003
C18:2 *cis*-9,*trans*-11	19	15	1.2	0.003
C18:2 *trans*-10,*cis*-12	1	0	0.1	NS
C18:3n-6	3	3	0.2	0.001
C18:3n-3	19	14	1.0	<0.001
C18:4n-3	1	1	0.1	0.002

[1]Nonsignificant = $P > 0.10$.

[2]Contrast of ORG vs. CON.
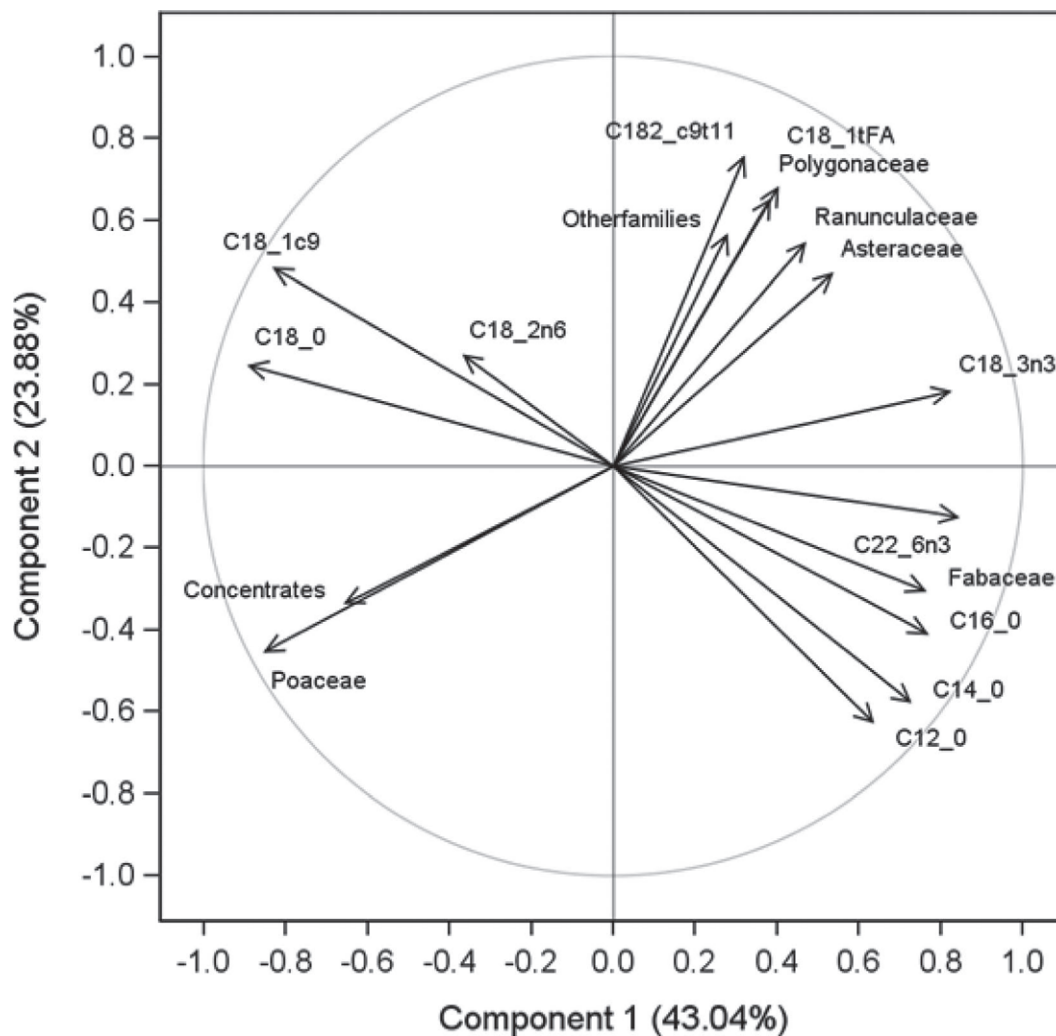
Figure 2. Correlation loading plot for first and second principal component (PC1 vs. PC2) showing the relationship between milk FA proportions of C12:0 (C12_0), C14:0 (C14_0), C16:0 (C16_0), C18:0 (C18_0), C18:1 *cis*-9 (C18_1c9), C18:1 *trans* FA (C18_1tFA), C18:2n-6 (C18_2n6), C18:2 *cis*-9,*trans*-11 (C18_2c9t11), C18:3n-3 (C18_3n3), C22:6n-3 (C22_6n3), and herbage proportions of the grass family (*Poaceae*), legume family (*Fabaceae*), knotweed family (*Polygonaceae*), aster family (*Asteraceae*), buttercup family (*Ranunculaceae*), other botanical families, and cows' concentrate DMI (means over 2 yr for 28 farms).

et al., 2004; Ellis et al., 2006; Collomb et al., 2008b). High intake of PUFA from fresh herbage affects rumen fermentation pattern and gives milk fat with less SFA and more C18:1 *trans*-11, C18:2 *cis*-9,*trans*-11, and C18:3n-3 compared with indoor feeding (Lock and Garnsworthy, 2003; Wiking et al., 2010). In contrast to previous findings (Elgersma et al., 2004; Ellis et al., 2006; Collomb et al., 2008b), the proportions of n-6 FA were higher in summer milk than in winter milk, and no seasonal changes in the n-6:n-3 FA ratios were observed in the present study. This is due to high milk fat proportions of C18:2n-6 in summer milk, but the underlying mechanisms are not known.

The negative correlations between milk fat proportions of C18:1 *trans* FA, C18:2 *cis*-9,*trans*-11, and grass herbage proportions (see Correlation of Herbage Botanical Composition and FA Composition in Milk) agree with the findings of Collomb et al. (2002). In the present study, legumes were positively correlated with C12:0, C14:0, and C16:0, which is in contrast with Collomb et al. (2002), who found a positive correlation between grass proportion and total SFA, and Dewhurst et al. (2003b), who found a negative effect of red clover proportion on milk fat proportion of C16:0.

Effects on Fat-Soluble Vitamins and Se in Milk and on Milk Sensory Quality

Milk concentrations of α-tocopherol and β-carotene were not affected by production system. These results are in accordance with Ellis et al. (2007) and Butler et al. (2008) in the indoor feeding period. In contrast to the present study, Butler et al. (2008) found positive effects of organic farming on α-tocopherol and β-carotene concentrations, and Ellis et al. (2007) found a negative effect on retinol concentrations in milk produced during the grazing season. These authors assumed that higher concentrations of α-tocopherol and β-carotene were related to grazing, intake of fresh forage, or intake of grass silage, whereas the effect on retinol most likely was due to higher vitamin A concentrations in the concentrates used on conventional farms than in those used on organic farms.

The milk Se concentrations in this study were in the range reported by others, but lower than reported in South Dakota, an area with naturally high soil Se concentrations (Alaejos and Romero, 1995; Lindmark-Månsson et al., 2003). The higher milk Se concentrations on ORG farms are most probably due to the inclusion of Se-rich fishmeal in the diets. Organically bound Se is more bioavailable than inorganic Se (selenite) and is translocated more efficiently to storage proteins and to milk; therefore, feeding organic Se results in higher milk Se concentrations than when feeding inorganic Se

(Calamari et al., 2010; Govasmark et al., 2010). Thus, the potentially higher amount of Se supplied and the higher Se bioavailability of the supplements may explain the higher Se concentration in milk produced on ORG farms.

In contrast to the studies of Ellis et al. (2007) and results from the outdoor feeding season in the study of Butler et al. (2008), we found no effects of production system on fat-soluble vitamins. Butler explained the differences during the outdoor feeding season by greater dietary contribution from grazing on organic farms compared with conventional high-input farms.

The higher concentrations of α-tocopherol, β-carotene and unchanged concentrations of retinol in milk from the indoor feeding periods compared with the outdoor feeding periods were unexpected and in contrast to other studies (Lindmark-Månsson et al., 2003; Agabriel et al., 2007). Concentrations of fat-soluble vitamins in silage decrease during storage (Beeckman et al., 2010), and therefore lower values in milk would be expected during the indoor feeding periods. This unexpected effect of season on vitamins warrants further study to clarify the mechanisms. Vitamin supplements were fed as synthetic vitamins on both ORG and CON farms, which likely explains the lack of differences between systems. However, lower milk yields on ORG farms may have resulted in increased vitamin concentrations, as reported by Jensen et al. (1999).

Elevated milk fat proportions of n-3 FA suggest positive health effects of organically produced milk for humans (Kliem and Givens, 2011); conversely, higher proportions of SFA are understood to have negative health effects. Although statistically significant effects of farming system were found for FA with potential health effects, for example, for C22:5 n-3, the absolute differences are most likely too small to have any effect on biological functions.

CONCLUSIONS

Despite differences in herbage botanical composition between ORG-SG and ORG-LG farms, grassland management had minor effects on milk FA composition. The effects of production system, organic versus conventional, on milk FA composition were pronounced and most likely related to differences in concentrate DMI and FA composition, although differences in herbage botanical composition existed between ORG farms and CON farms. Also, the effects of season were pronounced. Milk fat from ORG farms had higher proportions of health-beneficial n-3 FA, but also higher proportions of total SFA, which are regarded to have negative effects on health. Furthermore, the outdoor feeding periods with grazing had positive effects on

beneficial FA. Effects of grassland management and production system on fat-soluble vitamins and sensory quality were small.

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