

Authentication of Organic Milk Using $\delta^{13}\text{C}$ and the α -Linolenic Acid Content of Milk Fat

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The carbon stable isotope ratio ($\delta^{13}\text{C}$) and the α -linolenic acid (C18:3 ω 3) content of milk fat were analyzed to examine their applicability as general markers for the authentication of organic retail milk in Germany. To record the variable effect of feeding, including the seasonal influence on milk composition, three conventionally and three organically produced brands of retail milk were collected biweekly during a period of 18 months. Altogether 286 milk samples were analyzed. Threshold values for the identification of German organic milk were established and allowed to delimit almost all conventional samples. Organic retail milk was always above a minimum C18:3 ω 3 content of 0.50% and below a maximum $\delta^{13}\text{C}$ of -26.5‰ . The universal and strongly negative correlation ($r = -0.93$) between C18:3 ω 3 and $\delta^{13}\text{C}$ impedes the intentional manipulation of conventional milk. Conventional milk can naturally exceed the C18:3 ω 3 limit under atypical and rare conditions, but differentiation from organic milk can be improved by time-resolved comparison of data. In contrast with the general opinion, organic milk did not generally contain more c9,t11-C18:2 (CLA) than conventional milk. The proposed limits may deviate with dairy products containing milk from foreign countries.

KEYWORDS: Carbon stable isotopes; fatty acids; α -linolenic acid; authentication; organic milk

INTRODUCTION

The market for organic milk is still growing. In Germany, the sales of organic drinking milk increased by 34% in 2007 in comparison with the previous year, and its market share reached 11% for fresh drinking milk (1). In view of occasional shortages and a considerable price difference compared to conventional milk, there is a risk of wrongly labeled products. However, even comprehensive documentation for the purpose of traceability from the producer to the consumer cannot always prevent fraud. Besides economic reasons, consumers should be able to rely on the labeling for ecological reasons as well.

To protect the consumer from wrongly labeled dairy products, a laboratory method based on the analysis of product composition could deliver valuable additional information. Such a tool would not replace present controls along the process chain but would enable food-monitoring authorities to perform subsequent controls at the retail level whenever it was considered to be necessary.

Within the scope of our pilot study (2), that is, investigating the use of stable isotope analysis of carbon, nitrogen, and sulfur for the identification of organic milk, the $\delta^{13}\text{C}$ of milk fat proved to have good potential. Moreover, fatty acid analysis provided useful information on organic production in terms of the α -linolenic acid (C18:3 ω 3) content (2). Despite the seasonal variation of the $\delta^{13}\text{C}$ and C18:3 ω 3 content in milk fat, both

parameters showed a characteristic all-year difference between organic and conventional milk and allowed for the complete differentiation of the 35 samples analyzed.

The stable isotope ratio of nitrogen ($\delta^{15}\text{N}$) in plant material has been shown to reflect differences in the $\delta^{15}\text{N}$ of the fertilizer applied (3, 4). Because organic manure primarily has a higher $\delta^{15}\text{N}$ than artificial fertilizers, which are not allowed in organic production, isotope analysis has successfully been used to identify organically grown plant products under controlled conditions (5, 6). Although a specific organic N signature in feed plants should also be reflected in milk (7), the identification of organic milk by $\delta^{15}\text{N}$ could not be confirmed (2, 8). A possible explanation is the prevalent use of liquid manure from animal husbandry that is also applied in conventional feed production. Furthermore, $\delta^{15}\text{N}$ in soils and plants is influenced by leguminosae, such as clover, which fixate atmospheric N_2 and are used in conventional as well as in organic production. The fixated nitrogen has a lower $\delta^{15}\text{N}$ than organic manure, which is similar to artificial fertilizers.

The intended differentiation between organic and conventional milk by $\delta^{13}\text{C}$ would be attributable to the different percentages of maize in the respective feed. Maize is a C_4 plant and uses a different biosynthetic pathway to fixate atmospheric CO_2 than do C_3 plants, which comprise almost all other feed plants. The C_4 pathway results in a stronger accumulation of the heavier isotope ^{13}C compared to C_3 plants and, thus, in a higher $\delta^{13}\text{C}$ in C_4 plant material. Milk was shown to rapidly reflect changes in the carbon isotope signature of the feed (9). Consequently, the proportion of C_3 to C_4 plants ingested determines the $\delta^{13}\text{C}$

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in milk. Because an elevated portion of maize in the ration is typical of conventional milk production, whereas pasture-derived feed prevails in organic milk production (2), substantially different $^{13}\text{C}/^{12}\text{C}$ ratios (expressed as $\delta^{13}\text{C}$) can occur in milk (2, 9, 10). Corresponding findings were also reported for beef or cattle hair (11–13).

Differences in the C18:3 ω 3 content of milk fat are strongly associated with the feeding regimen as well. The feed in organic milk production typically consists of a high portion of roughage during the whole year, with fresh grass in the summer and grass/clover silage or hay in the winter. Moreover, the use of concentrates is limited relative to conventional production. A significantly higher C18:3 ω 3 percentage has repeatedly been reported for organic milk fat (2, 14–17). Such elevated C18:3 ω 3 levels in milk fat have been related to the limited use of maize silage and concentrates in feed (18, 19), a typical feeding regimen in organic milk production. A larger amount of pasture relative to concentrates has also been reported to significantly increase the concentration of ω 3 fatty acids and to reduce the ω 6/ ω 3 ratio in beef (20, 21).

It has, however, been demonstrated that an increase of C18:3 ω 3 in milk lipids is independent of the amount of C18:3 ω 3 in the ingested feed (19). Rather, it was postulated that the elevated C18:3 ω 3 content of milk fat is related to the increased bypass of dietary C18:3 ω 3 through the rumen. A ruminal energy deficit or inhibition of the ruminal flora by secondary plant metabolites has been discussed as a possible cause (19, 22). A negative energy balance could also mobilize C18:3 ω 3 from ruminant adipose tissue.

The objective of this study was to further investigate the applicability and robustness of the $\delta^{13}\text{C}$ and the α -linolenic acid content of milk fat for the authentication of organic milk. Thus, the task was to record the variable effect of feeding including the seasonal influence on milk composition in both organic and conventional milk production in more detail. The work focused on dairy-processed retail milk because of its high market share and the elevated risk of fraud compared to direct sales in farm shops.

MATERIALS AND METHODS

Samples. Milk samples were collected between November 2005 and May 2007 in biweekly intervals to cover the seasonal variation. Pasteurized whole milk was obtained from retail stores in Kiel, Germany. On each sampling day, three conventionally and three organically produced brands of milk were purchased. These retail samples originated from milk collection areas in northern, western, and southern Germany, and all were processed in trustworthy and renowned dairies. In addition, pasteurized milk was obtained from a single organic farm, certified by Bioland (23) and located ca. 10 km northwest of Kiel, Germany. Milk fat was extracted from milk samples according to the method of Roese-Gottlieb (24). However, to avoid heat-induced changes in fat composition, the solvents were removed by rotary evaporation at a maximum water bath temperature of 45 °C instead of drying at 102 °C. Altogether, 286 samples were collected and subjected to further analysis. Prepared fat samples were stored at –18 °C until analysis.

Gas Chromatography of Fatty Acids. Fatty acid methyl esters (FAME) were obtained from the milk fat samples by transesterification with sodium methylate. For this purpose a mixture of 1200 μL of *n*-heptane, 300 μL of a 10% solution of fat in *n*-heptane, and 30 μL of a 2 mol/L solution of sodium methylate in methanol was shaken vigorously for 3 min (vortex mixer) and centrifuged (2 min at 2000 min^{-1}). The supernatant was injected directly into the GC using the “hot needle” technique. FAME analyses were performed with a CP-3800 gas chromatograph (Varian, Palo Alto, CA) equipped with a split injection port (1:50), FID, and a 50 m fused silica capillary column (i.d. = 0.25 mm) coated with a 0.20 μm film of CP-Sil 88 (Varian).

Hydrogen was used as carrier gas at a flow of 2.1 mL/min (116 kPa). Injector and detector temperatures were 255 °C. The oven temperature was maintained at 50 °C, isothermal, for 1 min, then programmed at 5 °C min^{-1} to 225 °C, which was held for 3 min isothermal, and then programmed at 1 °C min^{-1} to 237 °C. The injection volume was 1 μL of a 2% solution. Evaluation of chromatograms was performed with an Agilent ChemStation (Santa Clara, CA). Calibration of the major fatty acids was carried out using the reference milk fat CRM 164 (IRMM, Geel, Belgium). Fatty acids in the range from C4 to C24 were determined and calculated as weight percentage (g/100 g of fatty acids).

Isotope Ratio Mass Spectrometry (IRMS). *Sample Preparation.* For the isotopic analysis of carbon, 0.46 mg of milk fat was weighed into tin capsules. Wrapped samples were introduced into an elemental analyzer Flash EA 1112 (Thermo Fisher Scientific, Waltham, MA) using an AS200 autosampler (Thermo Fisher Scientific). Samples were combusted at an oven temperature of 1020 °C with an oxygen pulse (injection time = 3 s) in a reactor packed with chromium(III) oxide and silvered cobaltous oxide. In a second reactor packed with copper wire, the NO_x formed was reduced to N_2 at 680 °C. Subsequently, water was trapped with magnesium perchlorate. At a continuous helium flow rate of 90 mL min^{-1} , the reaction gases N_2 and CO_2 were separated on a GC column at 45 °C and transferred to the mass spectrometer by use of a ConFlo III interface (Thermo Fisher Scientific) in dilution mode. Although the milk fat samples did not contain any nitrogen, the EA setup was retained to remove nitrogen traces originating from the autosampler blank.

Stable Isotope Analysis and Calibration. Analysis of $^{13}\text{C}/^{12}\text{C}$ ratios was performed with a Delta^{plus} XL isotope ratio mass spectrometer (Thermo Fisher Scientific) using the software ISODAT 1.5 (Thermo Fisher Scientific). Isotope ratios are given in ‰ on a δ scale and refer to the international standard VPDB. Carbon δ values are calculated as follows:

$$\delta^{13}\text{C} (\text{‰}) = \frac{R_{\text{sample}} - R_{\text{standard}}}{R_{\text{standard}}} \times 1000 \quad R = \frac{c_{13\text{C}}}{c_{12\text{C}}} \quad (1)$$

The standard deviation of the measurements ($n = 9$) was $\leq 0.05\text{‰}$ using the reference gas carbon dioxide. To take into account any inhomogeneity and, thus, to obtain data representative of the material, mean values of three individual analyses of each sample were determined. The standard deviation of these individual analyses was $< 0.15\text{‰}$ with a median value of 0.03‰.

Sucrose (Merck, Darmstadt, Germany) was calibrated as working standard using the international standards IAEA-CH-6 ($\delta^{13}\text{C}_{\text{VPDB}} = -10.4\text{‰}$), IAEA-CH-7 ($\delta^{13}\text{C}_{\text{VPDB}} = -31.8\text{‰}$), and NBS 22 ($\delta^{13}\text{C}_{\text{VPDB}} = -29.8\text{‰}$). Although the official reference values of these standards have slightly changed recently, the previous figures are used to ensure consistency within the current study and with the preceding work (2). The working standard was analyzed regularly in each sequence to control the measurement repeatability and to calibrate the reference gas carbon dioxide (Air Liquide, Düsseldorf, Germany).

RESULTS AND DISCUSSION

The study was performed to record in more detail the variation of selected fatty acid and stable isotope parameters in conventional and organic milk. Thus, to fully cover the seasonal variation, the sampling period comprised 18 months with biweekly intervals. Three conventionally and three organically produced brands of retail milk were purchased on each sampling day to also consider the variation between different milk collection areas. Additionally, samples were taken from a single organic farm to demonstrate the variability of milk composition potentially occurring in small herds. Altogether, 286 whole milk samples were analyzed in this study. The different origins of the samples are specified in **Table 1**.

The results obtained for the fatty acid composition refer to the lipid fraction and do not represent the content in milk

Table 1. Origins of Analyzed German Milk Samples

production	source	milk collection area	certified by	acronym
conventional	retail	Schleswig—Holstein		CRM 1
	retail	Mecklenburg—Western Pomerania		CRM 2
	retail	Bavaria		CRM 3
organic	retail	North Rhine—Westphalia	Bioland, Demeter, Naturland	ORM 1
	retail	Schleswig—Holstein and Lower Saxony	Bioland	ORM 2
	retail	Mecklenburg—Western Pomerania	Bioland, Demeter	ORM 3
organic	farm	Schleswig—Holstein, Kiel region	Bioland	OFM

because organic whole milk is usually marketed with a higher fat content of at least 3.8%, instead of 3.5% in conventional milk. The seasonal course of the α -linolenic acid (C18:3 ω 3) content in milk fat of different origins is shown in **Figure 1**. There were slightly elevated C18:3 ω 3 contents during the summer in the conventional milk brands CRM 1 and CRM 2 and an even more distinct increase for CRM 3. Starting from a higher level, the organic brands (ORM) showed a considerable increase of C18:3 ω 3 from spring to summer and a decrease in autumn. The strongest summer increase was observed with the OFM milk.

On the one hand, the effects are related to the seasonal change in feed composition caused primarily by the temporary availability of fresh pasture. On the other hand, the basically different C18:3 ω 3 levels in organic and conventional milk are attributable to the particular feed composition in organic milk production (18, 19). Compared with conventional milk production, organic feeding involves a limited use of maize silage and concentrates and, thus, an increased amount of pasture relative to concentrates.

With regard to the differentiation between organic and conventional samples, **Figure 1** shows that organic milk always had a higher C18:3 ω 3 content if related to single sampling days. However, if the all-year variation of C18:3 ω 3 is considered, the ranges for organic and conventional milk overlap to a certain extent. This result is caused by the conventional milk CRM 3, which originates from a region in southern Germany located at the foothills of the Alps. Here, the conventional milk production is characterized by comparatively increased pasture feeding. Even higher C18:3 ω 3 contents have been reported for summer milk obtained from the Swiss highlands, which can even exceed the values typical of organic milk production (25–27).

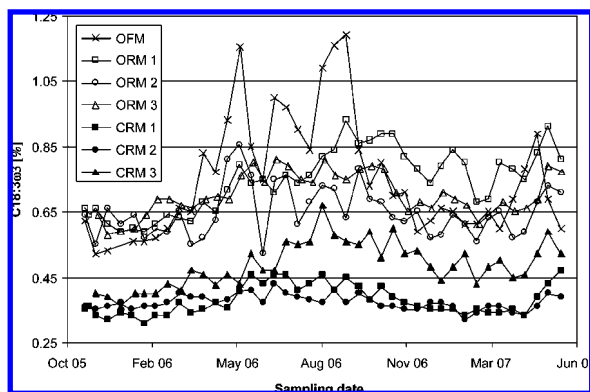


Figure 1. Seasonal variation of the C18:3 ω 3 content in milk fat (CRM, conventional retail milk; ORM, organic retail milk; OFM, organic farm milk).

Another aspect to be taken from **Figure 1** is the high variability of the C18:3 ω 3 content in organic milk obtained from a single farm (OFM). The examined farm, with a herd of 38 Holsteins, produced milk with the highest as well as with the almost lowest C18:3 ω 3 content found for organic milk in this study. Moreover, short-term variation between April and October 2006 was considerably increased compared with ORM 1 and ORM 3. A slight increase in the summer fluctuation of C18:3 ω 3 was established also for ORM 2, which is produced in a dairy collecting milk from only 17 farms. Short-term variation in the conventional milks CRM 1 and CRM 2 was comparatively low, particularly in the winter.

Generally, the short-term seasonal course of milk composition from a certain dairy is more balanced when more farms are included in the respective collection area. Thus, extreme values or short-term outliers of individual farms are largely leveled out in big dairies. Nevertheless, milk farms feeding an increased amount of pasture-derived feed relative to concentrates are more susceptible to fluctuations in the supply of self-produced feed caused by meteorological impact. In contrast to CRM 1 and CRM 2, which are largely based on high-performing farms with all-year barn feeding, CRM 3 and, even more so, ORM 1 showed a higher short-term variation of C18:3 ω 3. This is particularly obvious when the winter periods of 2006 and 2007 are compared.

Although there is an overlap between the all-year variation of C18:3 ω 3 in organic and conventional milk, the results nevertheless allow for the determination of a threshold value for organic milk. The minimum C18:3 ω 3 content found in organic milk fat was 0.52% in both retail and farm milk. Therefore, as a criterion for the authentication of organic milk, a C18:3 ω 3 content of >0.50% could be used. This limit would allow for the exclusion of the majority of conventionally produced German milk as nonorganic. Those conventional milk samples exceeding this limit usually are marketed at a higher price, just like the investigated CRM 3, and most likely will not be wrongly labeled organic.

Occasionally, dairies organize special feeding programs within the scope of conventional production (in the meaning of not being certified organic) aiming at producing a so-called pasture-milk. The thus obtained milk fat contains higher amounts of ω 3 fatty acids and conjugated linoleic acids (CLA). However, such milk is an independent niche product and will not be competitive compared to organic milk in terms of a lower price. Nevertheless, the vast majority of conventional milk production in Germany is performance-oriented, and conditions differ substantially from those of organic farming or similar feeding practices.

Previously, an elevated content of CLA in milk fat has been related to organic farming (14, 25). The finding that organic milk contains more CLA than conventional milk is generally accepted and used for marketing purposes. However, the results of this study do not confirm that as a general rule. **Figure 2** shows the seasonal course of the content of the predominant CLA isomer *c*9,*t*11-C18:2 in the analyzed milk fats. In winter, the highest CLA contents were established for CRM 3, which corresponds to the above-mentioned origin of this conventional milk. On the contrary, exceptionally low CLA contents of about 0.30% (minimum = 0.27%) were found for OFM in the winter. Furthermore, CRM 1 and CRM 2 showed higher CLA contents in the winter than ORM 3, whereas ORM 1 and ORM 2 were lower than CRM 3 most of the time.

CLA contents were higher and more variable during the summer in both organic and conventional milk, but not only

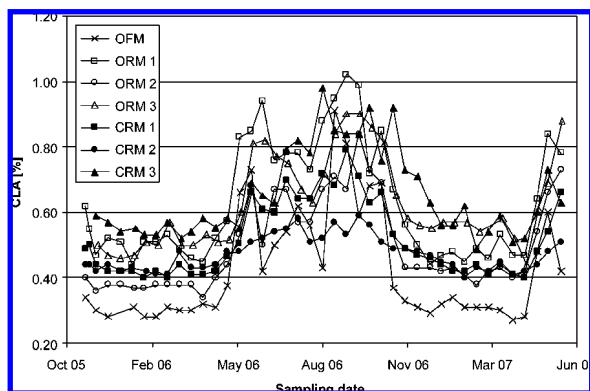


Figure 2. Seasonal variation of the CLA content (c9,t11-C18:2) in milk fat (see Figure 1 caption).

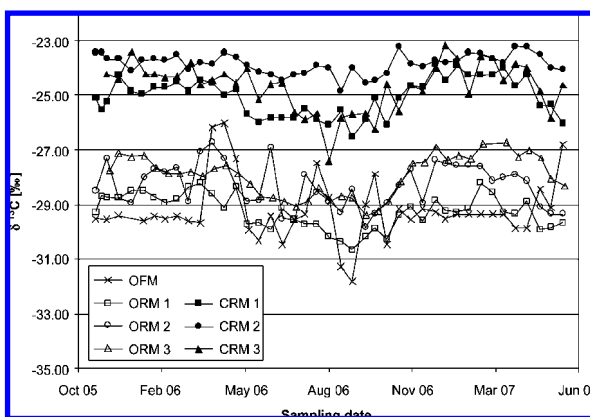


Figure 3. Seasonal variation of $\delta^{13}\text{C}$ in milk fat (see Figure 1 caption).

CRM 3 but even CRM 1 showed CLA contents within the variation range of 0.34–1.02% established for organic milk. The range for conventional milk was 0.40–0.98%. Thus, the CLA content is an obviously inappropriate criterion for differentiating organic from conventional milk. An increase of CLA is strongly associated with an increase in the *trans*-C18:1 (TFA) content (28). Thus, it was further confirmed in this study that TFA is an unsuitable marker for organic milk. Nevertheless, elevated contents of both CLA and TFA are indicative of pasture feeding.

The analysis of stable isotopes focused on carbon in this study because only the $\delta^{13}\text{C}$ of milk fat proved of good potential in our pilot study (2). Basically, a higher percentage of maize in the feed is reflected in a higher $\delta^{13}\text{C}$ of the resulting milk (9). The results obtained in this study are presented in Figure 3. The assumption that a different percentage of maize in the respective feed might generally be reflected in a distinctly different $\delta^{13}\text{C}$ of organic and conventional milk fat was evidently confirmed. All CRM samples had a clearly elevated $\delta^{13}\text{C}$ in comparison with ORM samples, giving evidence for the preferential use of maize in conventional milk production.

Even more distinct than the C18:3 ω 3 content (Figure 1), $\delta^{13}\text{C}$ shows a clear seasonal change with lower values in the summer for all milk brands (Figure 3). This effect demonstrates that, during the summer, the use of maize is decreased because of the increased availability of pasture feed in conventional as well as in organic milk production. However, the difference between both production systems is even higher than the respective within-system variation. Thus, with respect to the all-year variation there is no overlap between $\delta^{13}\text{C}$ in CRM and ORM samples except for one sample of CRM 3. Again, the single

Table 2. All-Year Variation of the C18:3 ω 3 Content and $\delta^{13}\text{C}$ in Milk Fat of Different Origins

series	n	18:3 ω 3 (%)				$\delta^{13}\text{C}$ (‰)			
		median	min	max	SD ^a	median	min	max	SD
CRM 1	42	0.36	0.31	0.47	0.05	-24.95	-26.52	-23.87	0.69
CRM 2	42	0.37	0.32	0.43	0.02	-23.78	-24.82	-23.20	0.37
CRM 3	40	0.48	0.36	0.67	0.07	-24.55	-27.42	-23.16	0.89
CRM total	124	0.39	0.31	0.67	0.07	-24.37	-27.42	-23.16	0.85
ORM 1	42	0.75	0.59	0.93	0.10	-29.24	-30.67	-28.19	0.64
ORM 2	41	0.64	0.52	0.86	0.08	-28.26	-29.84	-26.75	0.81
ORM 3	39	0.69	0.58	0.81	0.06	-27.86	-29.39	-26.71	0.74
ORM total	122	0.69	0.52	0.93	0.09	-28.63	-30.67	-26.71	0.92
OFM	40	0.70	0.52	1.19	0.18	-29.41	-31.82	-26.00	1.16

^a SD, standard deviation.

organic farm (OFM) showed the highest variation as well as extreme short-term jumps.

With these stable carbon isotope results, it appears rather practicable to define an upper threshold value for organic retail milk. The highest $\delta^{13}\text{C}$ found in ORM samples was -26.7‰. On the basis of this study, an exclusion criterion for organic milk of > -26.5 ‰ could be laid down. This would exclude $>99\%$ of the CRM samples. The only CRM sample lying below this limit originates from the CRM 3 series, which has already been described above as high-priced milk obtained under conditions of increased pasture feeding. This particular CRM 3 sample is identical to the one also showing the highest C18:3 ω 3 content of all CRM samples (Figure 1). Thus, the correct assignment of CRM 3 samples by $\delta^{13}\text{C}$ is by far less critical than by C18:3 ω 3.

In a recent Italian study (29), an extrapolated $\delta^{13}\text{C}$ maximum threshold value of -23.5‰ in casein was suggested for milk produced without using maize in the diet. According to that study, organic milk produced by feeding moderate amounts of maize, which in Germany occurs mainly in the winter, would have a slightly elevated $\delta^{13}\text{C}$ maximum in casein. As there is a variable depletion of ^{13}C during the synthesis of milk lipids compared to casein (29–31), the seeming inconsistency with the upper limit of -26.5‰ now established for organic milk lipids can be understood. However, the difference in $\delta^{13}\text{C}$ between casein and lipids depends on a variety of factors, such as the availability of lipids and protein from the feed and the portion of milk constituents derived from body material.

The variation ranges obtained for the different milk brands during the 18-month study are summarized in Table 2. Furthermore, the total ranges for CRM and ORM samples as well as for the single OFM series are given. Compared with our pilot study (2), the ranges for retail milk in Table 2 are extended for both C18:3 ω 3 content and $\delta^{13}\text{C}$, no longer allowing a 100% differentiation of organic samples from conventional ones. However, apart from still offering the possibility to define threshold values for organic milk, the present data also show further potential lying in the seasonal variation.

For both parameters, the seasonal course between the summer and winter was more or less parallel for organic and conventional milk, although each kind of milk varied on a different level. Thus, particularly the seasonal course of $\delta^{13}\text{C}$ (Figure 3) shows that at defined times there was a distinct difference between the lowest conventional and the highest organic milk.

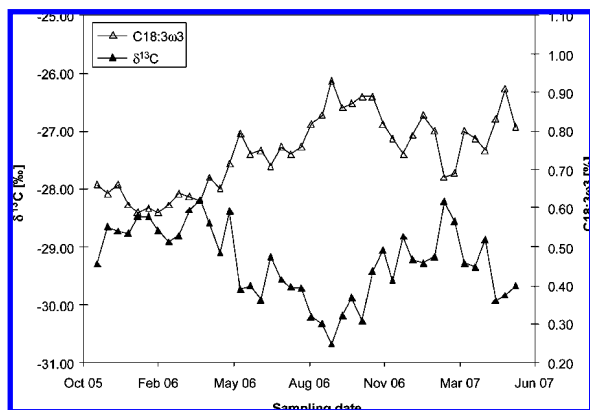


Figure 4. Seasonal variation of $\delta^{13}\text{C}$ and the C18:3 ω 3 content in ORM 1 milk fat.

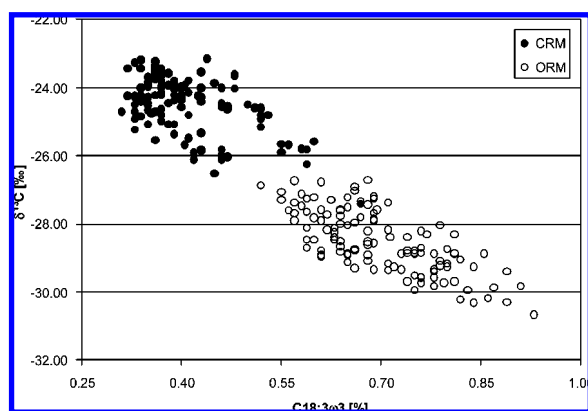


Figure 5. Correlation ($r = -0.93$) between the C18:3 ω 3 content and $\delta^{13}\text{C}$ of retail milk fat (see **Figure 1** caption).

This time-resolved difference between organic and conventional milk is rather constant and amounts to $2.64 \pm 0.55\text{‰}$ on average. Consequently, if the production date of a milk sample is known, the authentication of organic milk is substantially easier and more robust. The production date can be inferred from the “best before” date or be obtained exactly from the producer by quoting the batch number.

This effect is less distinct for the C18:3 ω 3 content (**Figure 1**), as the time-resolved average difference between organic and conventional milk is $0.16 \pm 0.07\text{‰}$ only. However, for the analyzed samples there was no overlap between the respective variations in both kinds of milk at a particular time.

Generally, there is a close relationship between the C18:3 ω 3 content and $\delta^{13}\text{C}$ of milk fat. This becomes apparent from **Figure 4**, which compares the seasonal course of both parameters in the ORM 3 samples. The different feeding effects on individual parameters have already been discussed in detail above. **Figure 4** demonstrates impressively that a low $\delta^{13}\text{C}$ is associated with a high C18:3 ω 3 content, and vice versa. The simple explanation is that an increase in the amount of pasture feed is accompanied by a decrease in the amount of maize.

This connection is not only valid for organic milk but can also be confirmed for all of the retail milk samples ($n = 246$) analyzed in this study. **Figure 5** shows a close correlation ($r = -0.93$) between the $\delta^{13}\text{C}$ and C18:3 ω 3 content of milk fat and, thus, demonstrates the general association of both parameters. The corresponding correlation established in our pilot study was $r = -0.95$. However, it was based upon 20 retail milk samples only (2).

Despite the close association between the C18:3 ω 3 content and $\delta^{13}\text{C}$ of milk fat, each is obtained by an independent

analytical method. However, from a practical point of view, it is advantageous that both parameters are measured in the easily accessible milk fat. The combination of two different parameters makes the authentication of organic milk more robust against intentional manipulation of conventional milk for the purpose of a faulty classification. In particular, this is favorable with respect to the possible use of suitable feed additives. One method for conventional milk producers to obtain a milk fat composition equivalent to organic milk would be using a feed composition and amount identical to those used by organic farmers. Because this approach involves a lower milk yield and, thus, increased production costs, there would hardly be an economic benefit to mislabel such milk as organic.

However, another approach to fraud would be the adulteration of organic milk by the addition of a certain percentage of conventional milk, in particular on the level of the processing dairy. Then, a fairly sensitive detection limit of 9.5% added conventional milk would only result if the C18:3 ω 3 content was at the lower limit (**Table 2**) in both the unadulterated organic and the added conventional milk. On the basis of the respective upper limits of $\delta^{13}\text{C}$, a minimum detection level of 5.9% conventional milk can be calculated. However, on the basis of a median composition (**Table 2**) of both milk fractions, the proposed threshold values allow for a detection of added conventional milk only above a level of 50% ($\delta^{13}\text{C}$) or even 63% (C18:3 ω 3). Thus, the data obtained in this study primarily are applicable to the differentiation of compositionally unadulterated milk.

Furthermore, the presented data principally characterize the composition of retail milk produced in Germany. Consequently, the derived threshold values for C18:3 ω 3 and $\delta^{13}\text{C}$ in organic milk—with the restriction of exceptionally occurring false-positive conventional samples—may be used for drinking milk made from German raw milk or milk produced under similar conditions in a foreign country. However, if the proposed compositional parameters are used with milk originating from other countries, the threshold values would possibly have to be checked or redetermined. Moreover, because $\delta^{13}\text{C}$ is linked to the use of maize, this parameter basically cannot be applied with conventional milk from countries not using maize as a forage plant.

Because, in contrast to drinking milk, certain processed dairy products may contain considerable amounts of milk components originating from foreign countries, the established threshold values may not always be applicable to German dairy products other than milk. In the context of this study, this concerns mainly the milk lipids. However, the composition of milk lipids can also be affected or changed during the processing of dairy products. Consequently, the authentication of processed dairy foods should be carried out with special care.

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