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Effects of milk type, production month, and brand on fatty acid composition: A case study in Korea

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ABSTRACT

The aim of this study was to examine the difference in fatty acid (FA) composition of organic and conventional milk at the retail market level in Korea for different milk production months and brands. The essential FA contents of the milk vary significantly under the combined effects of milk type, production month, and brand. Chemometric analysis reveals a greater difference between milk types than between production months and identifies significantly different levels of nutritionally desirable FAs—notably C18:3 *n*−3, C18:2 *n*−6 c and t—in the organic and conventional milks. Notwithstanding the limited sampling size and period, the results from this study may provide a better understanding of the nutritional quality of organic milk to consumers who are interested in organic milk intake.

1. Introduction

Milk is widely considered and consumed as a valuable source of important nutrients such as proteins, fats, and carbohydrates, as well as providing several physiologically functional compounds including bioactive peptides, antioxidants, essential vitamins/minerals, and nutritionally desirable fatty acids, notably omega-3 fatty acids (*n*−3 FAs). (Haug, Høstmark, & Harstad, 2007; Mills, Ross, Hill, Fitzgerald, & Stanton, 2011) For example, milk is a major source of fat in the human diet, accounting for 18–24%, 30–40%, and 20–25% respectively of total fat, saturated FA (SFA), and trans FA intake (Hulshof et al., 1999; Kliem, Shingfield, Livingstone, & Givens, 2013).

In Korea, the annual raw milk production was about 2.2 megatons in 2014, ~30% of which was used to prepare other dairy products such as cheese or yogurt (Korea_Dairy_Committee, 2014). In particular, owing to increased health awareness and the current drive towards improved quality of life, organic milk has been gaining interest in many countries. For example, organic milk already accounts for ~10% of the total milk market in Austria and Switzerland, and in spite of the price premium compared with conventional milk, the demand for organic milk in the US, the UK, and Germany is ever increasing (Molkentin & Giesemann, 2007). In general, consumers interested in organic products (such as organic milk) associate them with a higher nutritional value and are therefore willing to pay a premium price. However, whether the nutritional quality of organic products is actually superior is a rather controversial question whose answer may vary between foodstuffs (Bahar et al., 2008; Molkentin, 2013; Samman et al., 2008).

In general, organic farming practice aims to realize ecologically and/or socially sustainable natural systems by enhancing biodiversity, biological cycles, and soil biological activity. Although there are no worldwide standards and regulations governing organic agricultural products, governmental regulation bodies and some international organizations, such as the International Federation of Organic Agriculture Movements, have been established to harmonize organic production, labelling, and certification methods (Samman et al., 2008). The rules for organic milk farming are similar in many countries. For example, all feedstuffs used should be sourced organically and be free of antibiotics, synthetic fertilizers, and pesticides (O'Donnell, Spatny, Vicini, & Bauman, 2010). According to European standards for organic farming, animals should have sufficient access to an outdoor area and their daily feed ration should contain at least 60% roughage (Capuano et al., 2014). In addition, the use of concentrates is also limited and the feed must include a high proportion of roughage throughout the year. Cereal supplementation is allowed but only as a minor component to enhance milk production during winter when fresh dairy feedstuffs such as grass are scarce. More fresh pasture or fresh

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grass is therefore required for organic than for conventional milk production. Furthermore, in the winter, while organic milk production relies on a sufficient supply of (organic) hay, straw, or whole-crop (or grass) silage (Molkentin & Giesemann, 2007), the efficiency of conventional milk production is commonly enhanced in integrated dairy farming systems using concentrate, cereal, and/or (conserved) silage (Boner & Forstel, 2004). More detailed information on organic milk production standards can be found in European Union regulation No. 2092/91 (EU, 2004).

Regardless of previous conflicting reports, (Bergamo, Fedele, Iannibelli, & Marzillo, 2003; Toledo, Andrén, & Björck, 2002) organic milk is generally considered to contain higher amounts of nutritionally desirable components than conventional milk. Previous studies have indeed shown that organic bovine milk is richer in nutritionally desirable FAs, tocopherols, and/or carotenoids (Butler, Stergiadis, Seal, Eyre, & Leifert, 2011; Butler et al., 2008; Slots et al., 2009). However, there are health concerns related to the high SFA (i.e., lauric acid, myristic acid and/or palmitic acid) content of both organic and conventional milk, which has been associated with an increased risk of cardiovascular disease and metabolic syndromes in consumers (Butler et al., 2011; Haug et al., 2007).

The nutritional differences reported in Europe and the US between organic and conventional milk, notably in terms of FA composition, have been associated with several factors including dairy farming management systems, the milking season, sampling periods, and milk brands (Benbrook, Butler, Latif, Leifert, & Davis, 2013; Butler et al., 2011; O'Donnell et al., 2010). To our knowledge, although the volume of the organic milk market in Korea has been increasing by more than 65% each year (Korea_Dairy_Committee, 2014), nutritional differences between organic and conventional milk produced in Korea have yet to be characterized. This study therefore compares the FA contents of organic and conventional milks sold at the retail market level. The 37 FA in milk were profiled with gas chromatography coupled with a flame-ionization detector (GC–FID), and then statistically analyzed with chemometric approaches in order to reveal the milk FA variation depending on milk type, production month, and brand. The preliminary results from this study may provide a better understanding of the nutritional quality of organic milk to consumers who are interested in organic milk intake.

2. Materials and methods

2.1. Chemicals

Analytical or high-performance liquid chromatography grade methanol (MeOH), benzene, heptane, dimethoxypropane (DMP), and sulfuric acid (H_2SO_4) were purchased from J. T. Baker (Phillipsburg, NJ, USA), Fisher Scientific Korea Ltd. (Seoul, Korea), or Daejung Chemical & Materials Co. (Gyeonggi-Do, Korea). The mixture of 37 standard fatty acid methyl esters (FAME, CRM47885) and pentadecanoic acid (P6125) used as an internal standard were purchased from Sigma–Aldrich Co. (St. Louis, USA).

2.2. Milk collection and sample preparation for fatty acid measurements

Three popular organic and conventional milk brands were chosen on the basis of our prior report on the determination of organic milk authenticity from carbon and nitrogen isotope ratios (Chung, Park, Yoon, Yang, & Kim, 2014). The organic and conventional milks of each brand were purchased monthly from 2 to 3 local markets (near Konkuk University) in Seoul, Korea over three months (May–July 2014). The milk samples, 1 L of each milk brand and type, were purchased in triplicate on the same date every month. The milk samples were then lyophilized at a temperature below −40 °C for 3 days (Freexeone 4.5, Labconco, Kansas City, MO, USA) before being analyzed in terms of their FA contents.

To convert the FAs in the milk samples to the FAMEs prior to GC–FID analysis, ~50 mg milk powder samples were placed in tubes with Teflon-lined caps and 1.0 mg pentadecanoic acid was added as an internal standard. For the lipid extraction and FAME production, 340 μL methylation solvent (MeOH, benzene, DMP, and H_2SO_4 at 39:20:5:2, v/v/v/v) and 200 μL heptane were added to the samples and the resulting mixture was gently stirred and extracted at 80 °C for 2 h. Thereafter, the samples were cooled down to room temperature and the FAME in the aliquot supernatant was analyzed (Garces & Mancha, 1993).

2.3. FAME analysis by GC–FID

Gas chromatography was performed using an Agilent GC 7890A (Agilent Co. Ltd, USA) system coupled to a FID. A capillary column (DB-Wax, 0.25 mm × 30 m, 0.25 μm, Agilent Co. Ltd, USA) was used to separate the 37 FAMEs. The injection volume was 1 μL in 1:20 split mode. The carried gas was helium at 35 mL/min. The inlet temperature was 250 °C. The temperature of the GC oven was programmed as follows: 50 °C for 1 min, 200 °C (25 °C min^{-1}) for 5 min, and 230 °C (3 °C min^{-1}) for 20 min. The temperature of the FID was set to 280 °C. The total analysis time per sample was 42 min. Fig. 1 shows representative chromatograms of the 37 FAME standard mixture (Fig. 1A), and of the conventional (Fig. 1B), and organic milk (Fig. 1C) of brand B collected in May 2014. Individual FAs in the sample aliquot were identified by comparing their retention times with that of the mixture of 37 standard FAs. The FA content of the samples was calculated using the analytical method specified in the Food Code issued by the Korean Food and Drug Administration (Korean_Food_and_Drug_Administration, 2015).

2.4. Statistical analysis

Statistical analyses were performed using the general linear model of the statistical analysis program SAS (version 9.2; SAS Institute Inc., Cary, NC, USA). The experimental design was completely randomized with samples collected and analyzed in triplicate. Least significant difference tests were performed at the 0.05 probability level. Besides, a number of chemometric approaches—notably principal component analysis (PCA) and partial least-squares discriminate analysis (PLS-DA)—were used to discriminate the organic and conventional milks according to their FA composition. These methods are well suited to the classification of large biological datasets and allow the patterns therein to be assessed visually, via the score and loading plots that are the outcome of PCA and PLS-DA. The quantification data acquired by GC–FID were subjected to PCA (SIMCA-P version 13.0; Umetrics, Umeå, Sweden) to evaluate differences among groups of multivariate data. The data file was scaled to unit variance before the multivariate analysis. The PCA and PLS-DA output consisted of score plots to visualize the contrast between different samples and loading plots to explain the cluster separation.

3. Results

3.1. Comparison of milk fatty acid composition and content by GC–FID

Table 1 shows the effects of milk type, production month, and brand on the FA composition and content of the samples. In this study, we examined the variation in the FA contents of three