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Genetic and environmental relationships of detailed milk fatty acids profile determined by gas chromatography in Brown Swiss cows

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ABSTRACT

The aim of this study was to characterize the profile of 47 fatty acids, including conjugated linoleic acid (CLA), 13 fatty acid groups, and 5 Δ^9-desaturation indices in milk samples from Brown Swiss cows. The genetic variation was assessed and the statistical relevance of the genetic background for each trait was evaluated using the Bayes factor test. The additive genetic, herd-date, and residual relationships were also estimated among all single fatty acids and groups of fatty acids. Individual milk samples were collected from 1,158 Italian Brown Swiss cows and a detailed analysis of fat percentages and milk fatty acid compositions was performed by gas chromatography. Bayesian animal models were used for (co)variance components estimation. Exploitable genetic variation was observed for most of the de novo synthesized fatty acids and saturated fatty acids, except for C4:0 and C6:0, whereas long-chain fatty acids and unsaturated fatty acids (including CLA) were mainly influenced by herd-date effects. Herd-date effect explained large portions of the total phenotypic variance for C18:2 *cis*-9,*cis*-12 (0.668), C18:3 *cis*-9,*cis*-12,*cis*-15 (0.631), and the biohydrogenation and elongation products of these fatty acids. The desaturation ratios showed higher heritability estimates than the individual fatty acids, except for CLA desaturation index (0.098). Among the medium-chain fatty acids, C12:0 had greater heritability than C14:0 (0.243 vs. 0.097, respectively). Both C14:0 and C16:0 showed negative additive genetic correlations with the main monounsaturated and polyunsaturated fatty acids of milk fat, suggesting that their synthesis in the mammary gland may be influenced by the presence of unsaturated fatty acids. No correlation was observed between C4:0 and the other short-chain fatty acids (except for C6:0), confirming the independence of C4:0 from de novo mammary fatty acid synthesis. Among the genetic correlations dealing with potentially beneficial fatty acids, C18:0 was positively correlated with vaccenic and rumenic acids and negatively with linoleic acid. Finally, fatty acids C6:0 through C14:0 showed relevant correlations due to unknown environmental effects, suggesting the potential existence of genetic variances in micro-environmental sensitivity. This study allowed us to acquire new knowledge about the genetic and the environmental relationships among fatty acids. Likewise, the existence of genetic variation for most of de novo synthetized fatty acids and saturated fatty acids was also observed. Overall, these results provide useful information to combine feeding with genetic selection strategies for obtaining a desirable milk fatty acids profile, depending on the origin of fatty acids in milk.

Key words: Brown Swiss, genetic parameters, Bayes factor, fatty acids, Δ^9-desaturation index

INTRODUCTION

Milk and dairy products are sources of energy, high-quality protein, fat, minerals, and vitamins (Demment and Allen, 2003; Rooke et al., 2010). Bovine milk contains around 3 to 5% fat, represented 95 to 98% by triglycerides, which are in turn composed of glycerol and fatty acids. The fatty acid component is composed of 50 to 70% SFA, 20 to 40% of MUFA, and 1 to 5% PUFA (Jensen, 2002). The fatty acid composition of milk influences the milk's nutritional value and technological properties and reflects the cow's metabolic status (Fleischer et al., 2001; Mulligan et al., 2006). Recent data also suggest that it may reflect the cow's CH_4 production, as moderate relationships have been found between CH_4 production and some fatty acids (Chilliard et al., 2009; Dijkstra et al., 2011; van Lingen et al., 2014). Various feeding, management, and breeding strategies can be applied to manipulate the fatty acids profiles of milk (Lock and Bauman, 2004; Shingfield et al., 2013) and of processed milk products (Cattani et al., 2014).

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The possibility of modifying milk fat content and fatty acids profile through cow nutrition may enable producers to respond to human health recommendations in the context of a balanced diet (Woods and Fearon, 2009; Poulsen et al., 2012; Shingfield et al., 2013). For this purpose, particular attention has been paid to the presence in ruminant products of some PUFA, the n6-to-n3 fatty acid ratio, and CLA. These latter are produced by rumen biohydrogenation of linoleic acid and by the metabolism of various tissues, including the mammary gland, adipose tissue, and muscle (Lock and Bauman, 2004). Dietary CLA are reported to promote beneficial health-related effects on body composition and also show anticarcinogenic, antiatherogenic, antidiabetogenic, and immune-modulating properties (Rainer and Heiss, 2004). Likewise, feed ingredients rich in linolenic acid were found to be effective in reducing the n6-to-n3 ratio toward values <5, which are considered to be safe for the consumer (Danneberger et al., 2013).

Genetic variability has been found to influence the presence of some fatty acids, mainly those originating from de novo mammary fatty acid synthesis (e.g., Stoop et al., 2008; Mele et al., 2009; Krag et al., 2013), and between-breed differences have been observed in milk fatty acid profiles (White et al., 2001; Kelsey et al., 2003; Maurice-Van Eijndhoven et al., 2011). Furthermore, high-level genetic correlations have been observed among fatty acid that share similar metabolic production pathways (Soyeurt et al., 2007; Stoop et al., 2008).

Due to the high cost of GC analysis, most previous studies have focused on a limited number of fatty acid in a small number of animals. More recently, researchers have investigated the use of Fourier-transform infrared (**FTIR**) spectrometry to inexpensively predict the fatty acid concentration of bovine milk (Soyeurt et al., 2006; De Marchi et al., 2011; Bastin et al., 2013). The existing studies have considered various breeds, but examined relatively few (6–11) fatty acids. Gas chromatography has some major advantages over FTIR, including higher precision and accuracy in measuring a large number of fatty acids, even those present at low concentrations in milk fat (Rutten et al., 2010). Few studies have used GC to examine how genetic and herd factors affect the milk profile of 23 to 26 fatty acid traits: namely Stoop et al. (2008), Garnsworthy et al. (2010), and Heck et al. (2012). Only Bilal et al. (2014) reported data on 33 fatty acids and also evaluated all the genetic parameters based on GC analysis for the Holstein Friesian breed. In Brown Swiss cows, the genetic parameters of only 4 fatty acid categories (i.e., SFA, unsaturated fatty acids, MUFA, and PUFA predicted by FTIR and expressed as the relative content in a kilogram of milk) were previously estimated (Tullo et al., 2014). Few studies are available to evaluate the genetic, herd and residual correlations among different fatty acids (Duchemin et al., 2013). A deeper knowledge about the genetic or the environmental relationships among fatty acid provides useful information to identify the best strategies for manipulating the milk fatty acid profile.

The aims of this study were (1) to use a large GC-based data set to characterize the profile of 47 individual fatty acids, 13 fatty acid groups, and 5 calculated desaturation indices in Brown Swiss cows; (2) to estimate the genetic variation and heritability of these traits and assess their statistical relevance using the Bayes factor (**BF**) test; (3) to quantify the between herd-date variation; and (4) to infer the additive genetic, herd-date, and residual relationships among all fatty acids and fatty acid categories.

MATERIALS AND METHODS

Animals and Milk Sampling

Milk samples were collected from 1,158 Brown Swiss cows from 85 herds (a maximum of 15 cows per herd) located in the Alpine province of Trento (Italy). Milk samples were collected once per cow during the evening milking. Each farm was sampled once. The milk samples (no preservative was added) were immediately refrigerated at 4°C and transferred to the Cheese-Making Laboratory of the Department of Agronomy, Food, Natural Resources, Animals and Environment of the University of Padova (Legnaro, Padova, Italy). Data on the cows and herds were provided by the Superbrown Consortium of Bolzano and Trento (Italy), and pedigree information was supplied by the Italian Brown Swiss Cattle Breeders Association (ANARB, Verona, Italy). We included cows with phenotypic records available for the investigated traits and all known ancestors. Each sampled cow had known ancestors for at least 4 generations and the pedigree file included 8,845 animals.

GC Analysis

Fatty acid methyl esters were prepared by the direct extraction and alkali catalyzed trans-methylation procedure previously described by Feng et al. (2004). Briefly, each milk sample was centrifuged at 5,000 × g for 30 min at 4°C to facilitate the separation of the fat at the surface. Thirty milligrams of fat was collected in a fresh amber vial, mixed with 3 mL of hexane and 0.3 mL of 2 M methanolic solution of KOH, and the mixture was incubated for 5 min at room temperature after the addition of 0.25 mg of $NaHSO_3 \times H_2O$. The samples were then centrifuged at 3,000 × g for 3 min at 4°C and