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ORIGINAL ARTICLE

Genetic parameters for milk fatty acids, milk yield and quality traits of a Holstein cattle population reared under tropical conditions

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Summary

Information about genetic parameters is essential for selection decisions and genetic evaluation. These estimates are population specific; however, there are few studies with dairy cattle populations reared under tropical and sub-tropical conditions. Thus, the aim was to obtain estimates of heritability and genetic correlations for milk yield and quality traits using pedigree and genomic information from a Holstein population maintained in a tropical environment. Phenotypic records (n = 36 457) of 4203 cows as well as the genotypes for 57 368 single nucleotide polymorphisms from 755 of these cows were used. Covariance components were estimated using the restricted maximum likelihood method under a mixed animal model, considering a pedigree-based relationship matrix or a combined pedigree-genomic matrix. High heritabilities (around 0.30) were estimated for lactose and protein content in milk whereas moderate values (between 0.19 and 0.26) were obtained for percentages of fat, saturated fatty acids and palmitic acid in milk. Genetic correlations ranging from −0.38 to −0.13 were determined between milk yield and composition traits. The smaller estimates compared to other similar studies can be due to poor environmental conditions, which may reduce genetic variability. These results highlight the importance in using genetic parameters estimated in the population under evaluation for selection decisions.

Introduction

Estimates of heritability and genetic correlations are essential for the design of animal breeding programs and for the prediction of selection response. These values have been reported for several important traits in dairy cattle production, such as milk yield, fat and protein yield, fat and protein percentage, somatic cells score and more recently, fatty acids content. Nevertheless, genetic parameters are population specific and studies involving populations under tropical and subtropical conditions are still rare. A tropical environment is characterized by a long hot season, intense radiant energy and high relative humidity, which can cause several changes in physiology, anatomy and behaviour of lactating cows in an effort to maintain the heat balance (Curtis 1983). Heat stress can reduce feed intake and activity and increase respiration rate, peripheral blood flow and sweating, with a resulting harmful effect on production (West 2003).

These challenging conditions also affect the genetic variation and, consequently, the prediction of breeding values. In Cienfuegos-Rivas *et al.* (1999), the variance components were 40% lower in a Mexican population than in US environments, with a significant rank change of sires. Likewise, Costa *et al.* (2000)

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observed a reduction in genetic variability in subtropical regions in comparison with temperate regions. Therefore, selection decisions will be only correct if they were based on the information from the actual population under selection.

Traditionally, genetic parameters are estimated based on pedigree data. Nowadays, with the wide availability of genomic information, the genetic similarity between relatives can be determined more accurately, taking into account deviations due to Mendelian sampling. The use of molecular information is most useful when pedigree information is unavailable and the sample size is limited (Krag *et al.* 2013). The genomic information can be used by replacing the traditional relationship matrix based on pedigree information by a genomic relationship matrix (VanRaden 2008), that uses exclusively single nucleotide polymorphism (SNP) information, or by a matrix that combines the genomic and pedigree relationship information (Aguilar *et al.* 2010), including animals with and without genotype records. Studies which included genomic information for the estimation of genetic parameters in dairy cattle have obtained an increase in the accuracy of the estimates (Veerkamp *et al.* 2011; Haile-Mariam *et al.* 2013; Krag *et al.* 2013).

Therefore, due to the meagre number of studies in this area, the aim of this study was to estimate genetic parameters for milk production traits, including fatty acid composition, using pedigree and genomic information in a Holstein cattle population reared under tropical conditions.

Material and methods

Phenotypes

Monthly records of milk yield (MY; kg), somatic cells count (SCC), fat percentage (FP, %), protein percentage (PP, %), lactose percentage (LP, %), and palmitic (C16:0), stearic (C18:0), oleic (C18:1), total saturated (SFA), unsaturated (UFA), monounsaturated (MUFA), and polyunsaturated (PUFA) fatty acids percentage in milk (%) were collected from first through sixth parity Holstein cows located on four Brazilian farms between May of 2012 and December of 2014. Initially, the number of cows per farm was 3323, 544, 348, and 1003. Three of these farms are located in São Paulo state, with mean temperatures varying from 9.1 to 29.4°C and annual rainfall around of 1500 mm; and one is located in the state of Paraná, characterized by temperatures between 8.2 and 27.6°C and annual rainfall of 1971 mm. Cows are milked three times a day with an automatic milking system, maintained in freestall barns, and they are fed a total mixed ration. The main components of the ration are corn silage, grass hay, cotton seed, soybean meal, soybean hulls, corn meal, citrus pulp, minerals and vitamins.

Milk components were determined by mid-infrared spectroscopy (Delta Instruments CombiScope™ Filter; Advanced Instruments, Inc., Norwood, MA, USA; Rodriguez *et al.* 2014). A spectra treatment was performed by the manufacturer itself for fatty acids determination, but no calibration equations were developed in this study. The mid-infrared spectroscopy method was validated using gas chromatography (Rodriguez *et al.* 2014). The correlations between the measurements of milk fatty acids obtained by both methods varied from 0.60 to 0.92 and a bias ranging from −8.65 to 6.91 g/100 g of fat was estimated by using the Bland–Altman test. Despite this discrepancy, 94% of the samples were included within the concordance limits of the Bland–Altman test, indicating that the methods produced the same pattern of milk fat composition. Therefore, they allow similar conclusions about the milk samples under evaluation. More details regarding this validation are presented in Rodriguez *et al.* (2014).

The SFA group considered content of butyric acid (C4:0), caproic acid (C6:0), caprylic acid (C8:0), capric acid (C10:0), undecanoic acid (C11:0), lauric acid (C12:0), tridecanoic acid (C13:0), myristic acid (C14:0), pentadecanoic acid (C15:0), palmitic acid (C16:0), margaric acid (C17:0), stearic acid (C18:0), non-adecylic acid (C19:0), trycosylic acid (C23:0), lignoceric acid (C24:0); the MUFA group included the myristoleic acid (C14:1), cis-10-pentadecenoic acid (C15:1), palmitoleic acid (C16:1), cis-10-heptadecenoic acid (C17:1), oleic acid (C18:1ω9), elaidic acid (C18:1ω9), vaccenic acid (C18:1ω7), non-adecenoic acid (C19:1), erucic acid (C22:1ω9) and nervonic acid (C24:1), whereas the PUFA group included the linoleic acid (C18:2ω6), linolenic acid (C18:3ω3), linoleic conjugated acid (c9,t11-CLA), dihomo-γ-linolenic acid (C20:3ω3), docosadienoic acid (C22:2) and the eicosapentaenoic acid (EPA, C20:5). The UFA group was the sum of MUFA and PUFA cited previously.

The individual fatty acids C16:0, C18:0 and C18:1 were studied because of their importance in milk fat and potential impact on human health. The palmitic acid (C16:0) and the oleic acid (C18:1) are the main saturated and monounsaturated fatty acid of milk, respectively, with a concentration of 8 g per litre of milk (Haug *et al.* 2007). Both C16:0 and C18:1 have been associated with the levels of blood cholesterol (Kris-Etherton *et al.* 1999; Mensink *et al.* 2003), and

the C18:1 has been related to protection against oxidative stressors, atheromatosis and cardiovascular diseases (Haug *et al.* 2007). The synthesis of the oleic acid is strongly dependent on the stearic acid content. About 40% of the stearic acid is desaturated to oleic acid by the delta-9 desaturase in the mammary gland, contributing to more than 50% of the oleic acid secreted into milk fat (Enjalbert *et al.* 1998, Chilliard *et al.* 2000).

Somatic cells count (SCC) was transformed into somatic cells score (SCS) using the formula $SCS = Log_2(SCC/100\ 000) + 3$ for data normalization (Aka & Shook 1980). Initially, the SCC in the milk samples varied from 1000 to 15 974 000 cells per millilitre of milk.

Data from animals without valid measurements or with measurements outside the acceptable range (mean ± 3 standard deviations); without calving date, lactation order and/or age information were deleted to remove database inconsistency. Records of animals with days in milk lower than five or higher than 305, age higher than 9 years, and lactation order higher than six were also excluded. The descriptive statistics of the remaining data are presented in Table 1. There were 2614, 1937, 1135, 565, 242 and 48 cows from first-, second-, third-, fourth-, fifth- and sixth-lactation, respectively (1188 cows had records from two or more lactations). The numbers of cows per farm were 2800, 518, 139, and 746. The number of measures per cow varied from one to 28 and the average of measures per lactation per cow was 5.57.

Table 1 Number of observations (N), mean, standard deviation (SD), coefficient of variation (CV, %), minimum (MIN) and maximum (MAX) obtained for milk yield (MY), somatic cell score (SCS), fat percentage (FP), protein percentage (PP), lactose percentage (LP), and palmitic (C16:0), stearic (C18:0), oleic (C18:1), total saturated (SFA), unsaturated (UFA), monounsaturated (MUFA), and polyunsaturated (PUFA) fatty acids content in milk

Trait	N	Mean	SD	CV	MIN	MAX
MY (kg/day)	30 430	34.20	10.076	29.5	5.00	64.00
SCS	34 162	3.06	2.257	73.8	-2.64	10.11
FP (%)	32 952	3.45	0.748	21.7	1.00	6.03
PP (%)	33 195	3.05	0.302	9.9	2.07	4.23
LP (%)	33 051	4.60	0.238	5.2	3.56	5.25
Fatty acids (%)						
C16:0	29 731	0.84	0.221	26.2	0.15	1.60
C18:0	29 556	0.61	0.156	25.6	0.10	1.13
C18:1	29 580	0.67	0.214	32.1	0.01	1.40
SFA	29 696	2.23	0.506	22.7	0.61	3.95
UFA	29 606	1.03	0.298	28.8	0.07	2.06
MUFA	29 599	0.87	0.257	30.8	0.04	1.76
PUFA	29 744	0.16	0.049	30.8	0.01	0.32

Contemporary groups (CG) were formed by the combination of calving season [dry (April to September) or rainy (October to March)], calving year based on start of calving season, farm and month of analysis information. CG containing fewer than five individuals were eliminated. The final data set included 36 457 records from 4203 cows distributed in 298 CG (Figure 1) and daughters of 226 sires.

Genotypes

Genotypes from 768 of the measured cows were obtained with Illumina Bovine LD BeadChip (Illumina, San Diego, CA, USA), which has 6909 SNP. These animals are daughters of 113 sires and they are represented in 249 CG. The DNA was extracted from hair root samples by using NucleoSpin Tissue® Kit (Macherey-Nagel GmbH & Co. KG, Düren, Germany).

Imputation was used to augment the genotype data set. The reference population adopted for this imputation was bulls, sires of these cows, genotyped with the Illumina BovineSNP50 (Illumina, San Diego, CA, USA) or GeneSeek Genomic Profiler HD platform (Neogen Agrigenomics, Lexington, KY, USA). The software findhap.f90 (VanRaden 2015) was used to impute to 60.671 SNP based on population and family information. During this stage, eleven samples with duplicated genotypes and/or incompatible parent-offspring relationship were excluded, leaving 757 samples in the database. This procedure was performed by the Animal Genomics and Improvement Laboratory (Agricultural Research Service, United States Department of Agriculture).

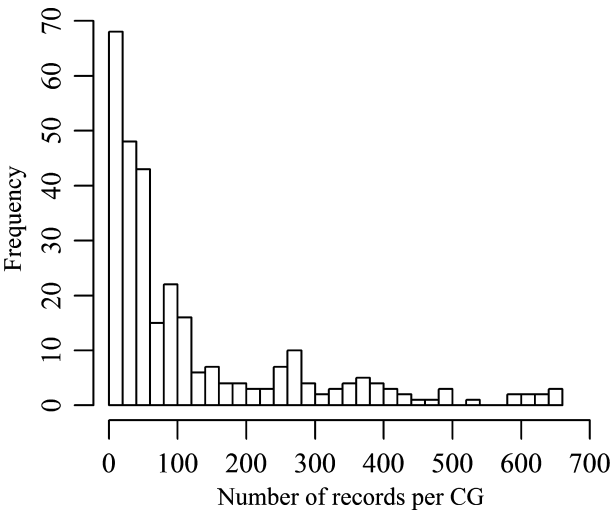

Figure 1 Distribution of records among contemporary groups (CG).

Samples with call rate lower than 90% (n = 2) as well as markers located in sex chromosomes (n = 1549), with proportion of missing genotypes higher than 20% (n = 145), monomorphic (n = 102), and with minor allele frequency lower than 0.02 (n = 1507) were also eliminated, leaving 755 cows and 57 368 SNP in the genetic analyses.

Genetic analyses

Genetic (co)variance components were estimated by using a numerator relationship matrix (pedigree-based approach) and by using a relationship matrix $\boldsymbol{H}$, which combined genomic and pedigree information (genomic and pedigree based approach). For both, the following univariate genetic model was fitted:

$$\mathbf{y} = \boldsymbol{X}\boldsymbol{\beta} + \boldsymbol{Z}\mathbf{a} + \boldsymbol{S}\mathrm{c} + \boldsymbol{\varepsilon} \quad (1)$$

where $\mathbf{y}$ is the vector of phenotypic observations; $\boldsymbol{X}$ is the design matrix for the fixed effects; $\boldsymbol{\beta}$ is the vector of fixed effects; $\boldsymbol{Z}$ is the design matrix for the additive genetic random effects; $\mathbf{a}$ is the vector of random additive genetic effects, which are the sum of genomic and residual polygenic effects; $\boldsymbol{S}$ is the design matrix for the permanent environmental random effects; $\mathbf{c}$ is the vector of permanent environmental random effects; and $\boldsymbol{\varepsilon}$ is the vector of residual effects, with $\boldsymbol{\varepsilon} \sim \mathrm{NID}\ (\mathbf{0}, \boldsymbol{I}\sigma^2)$. The vector $\boldsymbol{\beta}$ included the effects of CG, lactation order and the cubic effect of days in milk. Also, it was assumed $\mathbf{a} \sim \boldsymbol{N}\ (\mathbf{0}, \boldsymbol{A}\sigma_a^2)$ for the pedigree-based approach, and $\mathbf{a} \sim \boldsymbol{N}\ (\mathbf{0}, \boldsymbol{H}\sigma_a^2)$ for the combined approach.

The variance-covariance structure of genotyped animals in $\boldsymbol{H}$ is built from the genomic relationship matrix ($\boldsymbol{G}$) and the relationships of non-genotyped animals are adjusted in relation to the differences in genomic and pedigree-based relationships of their genotyped relatives (Koivula *et al.* 2012). This way, the inverse of the matrix $\boldsymbol{H}$ had the following structure (Aguilar *et al.* 2010):

$$\boldsymbol{H}^{-1} = \boldsymbol{A}^{-1} + \begin{bmatrix} \mathbf{0} & \mathbf{0} \\ \mathbf{0} & \boldsymbol{G}_w^{-1} - \boldsymbol{A}_{22}^{-1} \end{bmatrix}, \quad (2)$$

where $\boldsymbol{A}_{22}$ is the sub-matrix of $\boldsymbol{A}$ for genotyped animals, and $\boldsymbol{G}_w = w\boldsymbol{G} + (1 - w)\boldsymbol{A}_{22}$, with $\boldsymbol{G}$ equal to the genomic relationship matrix obtained by VanRaden (2008), and the constant w equal to 0.80, representing the proportion of the total additive genetic variance accounted by the genetic markers. The choice of w was based on Christensen *et al.* (2012), which found the optimal value of w as being between 0.70 and 0.85. The pedigree consisted of 8789 animals, 4197 dams and 576 sires of 6.85 generations, considering the first generation as one.

The phenotypic (r_p) and genetic (r_g) correlations between traits were estimated through the following bivariate model:

$$\begin{bmatrix} \mathbf{y}_1 \\ \mathbf{y}_2 \end{bmatrix} = \begin{bmatrix} \boldsymbol{X}_1 & \mathbf{0} \\ \mathbf{0} & \boldsymbol{X}_2 \end{bmatrix} \begin{bmatrix} \boldsymbol{\beta}_1 \\ \boldsymbol{\beta}_2 \end{bmatrix} + \begin{bmatrix} \boldsymbol{Z}_1 & \mathbf{0} \\ \mathbf{0} & \boldsymbol{Z}_2 \end{bmatrix} \begin{bmatrix} \mathbf{a}_1 \\ \mathbf{a}_2 \end{bmatrix} + \begin{bmatrix} \boldsymbol{S}_1 & \mathbf{0} \\ \mathbf{0} & \boldsymbol{S}_2 \end{bmatrix} \begin{bmatrix} \mathbf{c}_1 \\ \mathbf{c}_2 \end{bmatrix} + \begin{bmatrix} \boldsymbol{\varepsilon}_1 \\ \boldsymbol{\varepsilon}_2 \end{bmatrix} \quad (3)$$

where the vector $\mathbf{y_1}$ and $\mathbf{y_2}$ refer to the observations of the first and second traits, respectively; $\boldsymbol{X_1}$ and $\boldsymbol{X_2}$ are the design matrices and $\boldsymbol{\beta_1}$ and $\boldsymbol{\beta_2}$ are the vectors of the fixed effects for the first and second trait, respectively; $\boldsymbol{Z_1}$ and $\boldsymbol{Z_2}$ are the design matrices and $\mathbf{a_1}$ and $\mathbf{a_2}$ are the vectors of the random additive genetic effects; $\boldsymbol{S_1}$ and $\boldsymbol{S_2}$ are the design matrices and $\mathbf{c_1}$ and $\mathbf{c_2}$ are the vectors of random permanent environmental effects; and $\boldsymbol{\varepsilon_1}$ and $\boldsymbol{\varepsilon_2}$ are the vectors of the residual effects NID ($\mathbf{0}$, $\boldsymbol{I}\sigma_e^2$).

The covariance components were estimated using the restricted maximum likelihood method with an average information algorithm under models (1) and (3), using the software AIREMLF90 (Misztal *et al.* 2002). Standard errors for additive genetic, permanent environmental and residual variance and covariance components were computed as square roots of diagonal elements of the inverse of the average information matrix. Heritability (h^2) was calculated as the ratio of the additive genetic variance to the phenotypic variance whereas the proportion of phenotypic variance due to permanent environmental effects (c^2) was determined as the ratio of permanent environment variance to the phenotypic variance. For these functions of variance components as well as for the genetic and phenotypic correlations, standard deviations obtained from the repeated sampling approach were considered as their standard errors (Meyer & Houle 2013).

Results

The estimates of variance components and heritability for milk yield and composition traits obtained through pedigree-based and genomic-based approaches are given in Tables 2 and 3, respectively, whereas genetic and phenotypic correlations are shown in Tables 4 and 5. Small differences in these estimates (and their standard errors) were observed between the methods used, especially in the univariate analyses. The standard errors of genetic correlations were lower when

genotypes were used, with differences between 0.001 and 0.007. This reduction was more evident for the genetic correlations between SCS and the other traits, for which the standard errors varied from 0.086 to 0.122 and from 0.081 to 0.115 in pedigree and genomic-based approaches, respectively. In general, the additive variance and genetic correlations were slightly higher in genomic analyses.

High heritabilities were estimated for PP and LP, with values greater than 0.30. Moderate estimates were obtained for FP, SFA, and C16:0, whereas the other traits had low estimates of heritability, varying between 0.07 and 0.14. The effect of permanent environment was mainly important for MY and SCS, corresponding to approximately 0.30 of the phenotypic variance (Tables 2 and 3).

Negative and moderate genetic correlations were estimated between MY and milk components, presenting a stronger association with PP ($r_g = -0.45$ and -0.42) and FP ($r_g = -0.40$ and -0.39). SCS also presented negative correlations (in this case, favourable) with MY and milk composition traits, except with the unsaturated fatty acids (UFA, MUFA, PUFA, and C18:1); however, these correlations were low and with high standard errors.

Positive genetic associations were verified among the milk composition traits. The maximum correlation was observed between FP and C16:0 ($r_g = 0.98$); FP and SFA, SFA and C16:0, and UFA and MUFA ($r_g = 0.99$). The genetic correlations between FP and the unsaturated fatty acids studied were slightly lower, but still high, with values ranging from 0.50 to 0.80. As expected, the milk fatty acids were highly correlated with each other, with the lowest genetic correlation observed for PUFA and C18:0 ($r_g = 0.32$ and 0.29 from the two analyses).

Protein percentage displayed positive and moderate to high genetic correlations with the other milk components, varying between 0.31 and 0.69. In turn, lactose percentage was positively but weakly correlated with FP, SFA, C16:0, C18:0 and C18:1 ($r_g < 0.15$); and moderately correlated with UFA, MUFA, and PUFA; presenting genetic correlations around 0.25.

Regarding the phenotypic correlations, lower estimates were observed for PP and LP with other traits ($r_p < 0.22$). On the other hand, larger and positive estimates were obtained for FP and the studied fatty acids with other traits. As well as genetic correlation estimates, MY showed negative and low to moderate phenotypic correlations with milk composition traits, except LP ($r_p = 0.18$). In general, SCS presented lower and positive phenotypic associations with other traits, but moderate and negative with LP ($r_p = -0.35$).

Discussion

According to Misztal *et al.* (2013), the gain in accuracy in genetic prediction due to the addition of genomic information can be small because the mean difference between the genomic relationship and the relationship based on pedigree is generally low. This gain is even smaller when animals with low individual accuracy are used, such as animals with their own records only. However, the inclusion of cows' genotypes in genetic

Table 2 Variance components, heritability and proportion of the variance due to permanent environmental effects,* and their respective standard errors (in brackets) obtained through pedigree-based analyses for milk yield (MY), somatic cell score (SCS), fat percentage (FP), protein percentage (PP), lactose percentage (LP), total saturated (SFA), unsaturated (UFA), monounsaturated (MUFA), and polyunsaturated (PUFA) fatty acids, and palmitic acid (C16:0), stearic acid (C18:0), and oleic acid (C18:1) content in milk

Trait	$\hat{\sigma}_a^2$	$\hat{\sigma}_{pe}^2$	$\hat{\sigma}_e^2$	$\hat{\sigma}_p^2$	h^2	c^2
MY (kg/day)	8.424 (1.5036)	19.685 (1.3187)	40.475 (0.3545)	68.584 (0.8999)	0.12 (0.021)	0.29 (0.020)
SCS	0.520 (0.0876)	1.293 (0.0776)	2.482 (0.0204)	4.296 (0.0552)	0.12 (0.020)	0.30 (0.018)
FP (%)	0.096 (0.0097)	0.036 (0.0071)	0.381 (0.0032)	0.514 (0.0057)	0.19 (0.018)	0.07 (0.014)
PP (%)	0.023 (0.0018)	0.007 (0.0012)	0.036 (0.0003)	0.065 (0.0010)	0.35 (0.024)	0.10 (0.019)
LP (%)	0.013 (0.0012)	0.007 (0.0009)	0.024 (0.0002)	0.044 (0.0007)	0.30 (0.025)	0.15 (0.021)
Fatty acids (%)						
C16:0	0.0105 (0.00094)	0.0033 (0.00066)	0.0263 (0.00023)	0.0401 (0.00053)	0.26 (0.021)	0.08 (0.017)
C18:0	0.0024 (0.00029)	0.0013 (0.00023)	0.0143 (0.00013)	0.0180 (0.00019)	0.13 (0.016)	0.07 (0.013)
C18:1	0.0024 (0.00039)	0.0019 (0.00033)	0.0290 (0.00026)	0.0333 (0.00031)	0.07 (0.011)	0.06 (0.010)
SFA	0.0570 (0.00518)	0.0189 (0.00367)	0.1520 (0.00135)	0.2279 (0.00293)	0.25 (0.021)	0.08 (0.017)
UFA	0.0051 (0.00081)	0.0033 (0.00067)	0.0577 (0.00051)	0.0661 (0.00061)	0.08 (0.012)	0.05 (0.010)
MUFA	0.0035 (0.00058)	0.0025 (0.00048)	0.0430 (0.00038)	0.0490 (0.00045)	0.07 (0.012)	0.05 (0.010)
PUFA	0.0002 (0.00003)	0.0001 (0.00002)	0.0015 (0.00001)	0.0018 (0.00002)	0.11 (0.014)	0.07 (0.012)

*$\hat{\sigma}_a^2$ additive genetic variance, $\hat{\sigma}_{pe}^2$ permanent environment variance, $\hat{\sigma}_e^2$ residual variance, $\hat{\sigma}_p^2$ phenotypic variance, h^2 heritability, c^2 proportion of phenotypic variance due to permanent environmental effects.

Table 3 Variance components, heritability and proportion of the variance due to permanent environmental effects,* and their respective standard errors (in brackets) obtained through genomic analyses for milk yield (MY), somatic cell score (SCS), fat percentage (FP), protein percentage (PP), lactose percentage (LP), total saturated (SFA), unsaturated (UFA), monounsaturated (MUFA), and polyunsaturated (PUFA) fatty acids, and palmitic acid (C16:0), stearic acid (C18:0), and oleic acid (C18:1) content in milk

Trait	$\hat{\sigma}_a^2$	$\hat{\sigma}_{pe}^2$	$\hat{\sigma}_e^2$	$\hat{\sigma}_p^2$	h^2	c^2
MY (kg/day)	8.589 (1.5098)	19.528 (1.3197)	40.475 (0.3545)	68.591 (0.9010)	0.13 (0.021)	0.28 (0.020)
SCS	0.536 (0.0884)	1.279 (0.0779)	2.482 (0.0204)	4.297 (0.0553)	0.12 (0.020)	0.30 (0.018)
FP (%)	0.097 (0.0096)	0.035 (0.0070)	0.381 (0.0032)	0.514 (0.0057)	0.19 (0.018)	0.07 (0.014)
PP (%)	0.023 (0.0018)	0.006 (0.0012)	0.036 (0.0003)	0.065 (0.0010)	0.35 (0.023)	0.09 (0.019)
LP (%)	0.014 (0.0012)	0.006 (0.0009)	0.024 (0.0002)	0.044 (0.0007)	0.31 (0.025)	0.14 (0.021)
Fatty acids (%)						
C16:0	0.0106 (0.00093)	0.0032 (0.00065)	0.0263 (0.00023)	0.0401 (0.00053)	0.26 (0.021)	0.08 (0.017)
C18:0	0.0024 (0.00029)	0.0013 (0.00023)	0.0143 (0.00013)	0.0180 (0.00019)	0.14 (0.016)	0.07 (0.013)
C18:1	0.0025 (0.00039)	0.0018 (0.00033)	0.0290 (0.00026)	0.0333 (0.00031)	0.07 (0.011)	0.06 (0.010)
SFA	0.0576 (0.00515)	0.0183 (0.00364)	0.1520 (0.00135)	0.2279 (0.00293)	0.25 (0.021)	0.08 (0.016)
UFA	0.0053 (0.00081)	0.0031 (0.00066)	0.0577 (0.00051)	0.0661 (0.00061)	0.08 (0.012)	0.05 (0.010)
MUFA	0.0036 (0.00058)	0.0024 (0.00048)	0.0430 (0.00038)	0.0490 (0.00045)	0.07 (0.012)	0.05 (0.010)
PUFA	0.0002 (0.00003)	0.0001 (0.00002)	0.0015 (0.00001)	0.0018 (0.00002)	0.11 (0.014)	0.06 (0.012)

*$\hat{\sigma}_a^2$ additive genetic variance, $\hat{\sigma}_{pe}^2$ permanent environment variance, $\hat{\sigma}_e^2$ residual variance, $\hat{\sigma}_p^2$ phenotypic variance, h^2 heritability, c^2 proportion of phenotypic variance due to permanent environmental effects.

Table 4 Phenotypic correlations (above diagonal), genetic correlations (below diagonal) obtained through pedigree-based analyses for milk yield (MY), somatic cell score (SCS), fat percentage (FP), protein percentage (PP), lactose percentage (LP), total saturated (SFA), unsaturated (UFA), monounsaturated (MUFA), and polyunsaturated (PUFA) fatty acids, and palmitic acid (C16:0), stearic acid (C18:0), and oleic acid (C18:1) content in milk*

	MY	SCS	FP	PP	LP	SFA	UFA	MUFA	PUFA	C16:0	C18:0	C18:1
MY		−0.24	−0.06	−0.26	0.18	−0.06	−0.04	−0.03	−0.03	−0.09	−0.06	−0.01
SCS	−0.13		0.04	0.20	−0.35	0.02	0.06	0.06	0.07	0.06	0.02	0.01
FP	−0.40	0.00		0.16	0.02	0.95	0.83	0.83	0.71	0.90	0.82	0.81
PP	−0.45	−0.04	0.55		0.04	0.15	0.10	0.08	0.22	0.13	0.05	0.03
LP	−0.21	−0.22	0.14	0.39		0.03	0.01	0.00	0.00	0.01	0.01	0.01
SFA	−0.36	−0.08	0.99	0.47	0.13		0.70	0.70	0.59	0.96	0.78	0.68
UFA	−0.34	0.11	0.78	0.55	0.26	0.66		1.00	0.87	0.62	0.80	0.97
MUFA	−0.32	0.09	0.79	0.50	0.26	0.68	0.99		0.82	0.64	0.79	0.98
PUFA	−0.38	0.19	0.52	0.69	0.25	0.39	0.79	0.70		0.44	0.70	0.77
C16:0	−0.35	−0.13	0.98	0.40	0.15	0.99	0.67	0.69	0.36		0.65	0.62
C18:0	−0.28	−0.07	0.86	0.33	0.12	0.85	0.66	0.70	0.32	0.82		0.78
C18:1	−0.29	0.03	0.80	0.42	0.15	0.71	0.93	0.95	0.60	0.71	0.77	

*Standard errors of genetic correlations varied from 0.002 to 0.122 whereas the standard errors of phenotypic correlations varied from 0.000 to 0.010.

evaluation is important to control a possible bias due to selection (Patry & Ducrocq 2011). Also, these genotypes can be used to correct pedigree errors. Tsuruta *et al.* (2013) reported an increase of 2–3% in the reliability of genomic breeding values for US final score through the inclusion of Holstein cows' genotypes in the genetic evaluation. In Ding *et al.* (2013), the use of 3087 cows genotyped for 48 676 SNP as a reference population resulted in accuracies between 0.70 and 0.80 in a validation population formed by 67 proven sires.

Nevertheless, in relation to genetic parameters, few studies assessed the impact of genomic information on their estimation. Veerkamp *et al.* (2011) estimated genetic parameters for milk yield, body weight and dry matter intake using relationships between animals based on pedigree, 43 011 SNP, or a combination of these in a dataset formed by 639 (517 with genotypes) primiparous Holstein cows. They observed a reduction in heritability estimates when genomic relationships were used, possibly due to base and scale differences between pedigree and genomic-based matrices and the additional information on the Mendelian sampling component in the genomic matrix. However, the use of SNP-based relationships improved the precision of these estimates. In a similar study of

Table 5 Phenotypic correlations (above diagonal), genetic correlations (below diagonal) obtained through genomic analyses for milk yield (MY), somatic cell score (SCS), fat percentage (FP), protein percentage (PP), lactose percentage (LP), total saturated (SFA), unsaturated (UFA), monounsaturated (MUFA), and polyunsaturated (PUFA) fatty acids, and palmitic acid (C16:0), stearic acid (C18:0), and oleic acid (C18:1) content in milk*

	MY	SCS	FP	PP	LP	SFA	UFA	MUFA	PUFA	C16:0	C18:0	C18:1
MY		−0.24	−0.06	−0.26	0.18	−0.05	−0.03	−0.03	−0.02	−0.09	−0.06	−0.01
SCS	−0.14		0.04	0.20	−0.35	0.02	0.06	0.05	0.07	0.05	0.02	0.01
FP	−0.39	−0.03		0.15	0.01	0.95	0.83	0.83	0.71	0.90	0.82	0.81
PP	−0.42	−0.04	0.52		0.04	0.15	0.10	0.07	0.22	0.13	0.05	0.03
LP	−0.20	−0.27	0.12	0.35		0.02	0.00	0.00	0.00	0.00	0.01	0.00
SFA	−0.35	−0.10	0.99	0.45	0.12		0.70	0.70	0.59	0.96	0.78	0.68
UFA	−0.29	0.04	0.77	0.53	0.22	0.65		1.00	0.87	0.62	0.80	0.97
MUFA	−0.28	0.02	0.78	0.48	0.22	0.67	0.99		0.82	0.64	0.79	0.98
PUFA	−0.33	0.13	0.50	0.68	0.23	0.36	0.78	0.69		0.43	0.70	0.77
C16:0	−0.34	−0.13	0.98	0.38	0.13	0.99	0.65	0.68	0.33		0.65	0.62
C18:0	−0.27	−0.11	0.86	0.31	0.11	0.84	0.64	0.67	0.29	0.82		0.78
C18:1	−0.27	0.02	0.80	0.40	0.12	0.70	0.93	0.95	0.59	0.70	0.76	

*Standard errors of genetic correlations varied from 0.002 to 0.118 whereas the standard errors of phenotypic correlations varied from 0.000 to 0.010.

Haile-Mariam *et al.* (2013), considering daughter trait deviations (DTD) as phenotypes of 2216 Holstein bulls genotyped for 45 993 SNP, a lower fraction of the variation in DTD was explained when using the genomic matrix in comparison to the pedigree-based matrix. This lower 'heritability' was attributed to a possible imperfect linkage disequilibrium between the SNP and the quantitative trait loci (QTL) influencing the trait.

Herein, the estimates of heritability and correlation obtained by using pedigree-based and combined relationship matrices were similar, probably due to the resemblance between these matrices, as argued by Misztal *et al.* (2013). The mean difference among the elements in the inverse of $\boldsymbol{A}$ and $\boldsymbol{H}$ was only 0.01 whereas the mean difference between $\boldsymbol{G}$ (genomic matrix) and $\boldsymbol{A_{22}}$ (sub-matrix of $\boldsymbol{A}$ for genotyped animals) matrices was equal to 0.0000024 (Figure 2). Furthermore, the amount of information per cow may have also minimized the impact of the genomic data on the results. The number of monthly records per cow varied from one to 28. In addition, 1010 cows with phenotypes also have daughters measured. It must be recognized that the main advantage of genomics is to provide an accurate evaluation of animals at birth. Therefore, although genomics can add accuracy to cows with their own data, in such case little was left for the genotypes to explain and, consequently, it was possible to obtain reliable estimates without SNP information. A greater difference between traditional and genomics approaches would be probably observed if the number of genotyped animals was higher and if bulls' genotypes were included in the analyses. However, the slightly lower standard errors obtained for the genetic correlations in the genomic approach confirmed this method as a useful tool to increase the accuracy of estimates.

In general, the estimates of heritability were lower than those estimated in Soyeurt *et al.* (2008), Stoop *et al.* (2008), Schopen *et al.* (2009), and Bastin *et al.* (2011), which also used measurements determined by mid-infrared spectrometry. The heritability for FP in these studies varied from 0.37 to 0.50 whereas for PP ranged from 0.44 to 0.66. This difference can be due to the use of first-parity cows. Differences in heritabilities for milk traits across parities were reported by Bastin *et al.* (2013), with generally higher heritabilities estimated in first parity than in later parities. Walsh *et al.* (2007), based on McDonald (1968), attributed the effect of parity on milk characteristics to longer and more dilated streak canals presented by multiparous cows in comparison to primiparous cows. Also, in Stoop *et al.* (2008) and Schopen *et al.* (2009), the cows were between 63 and 282 days in milk, a period in which higher heritabilities for these two traits were reported (Bastin *et al.* 2011). However, Penasa *et al.* (2015) obtained estimates similar to those of the present study. The estimates of heritability for FP and PP were 0.201 and 0.267, respectively, in a population of 25 317 multiparous Holstein cows with milk samples collected between five and 365 days in milk.

The same behaviour was observed for LP, with estimates of heritability ranging from 0.478 and 0.508 in Miglior *et al.* (2007) and equal to 0.62 in Schopen *et al.* (2009). However, a closer estimate of 0.33 for LP was estimated by Tiezzi *et al.* (2013) considering monthly records of 25 590 Holstein-Friesian cows

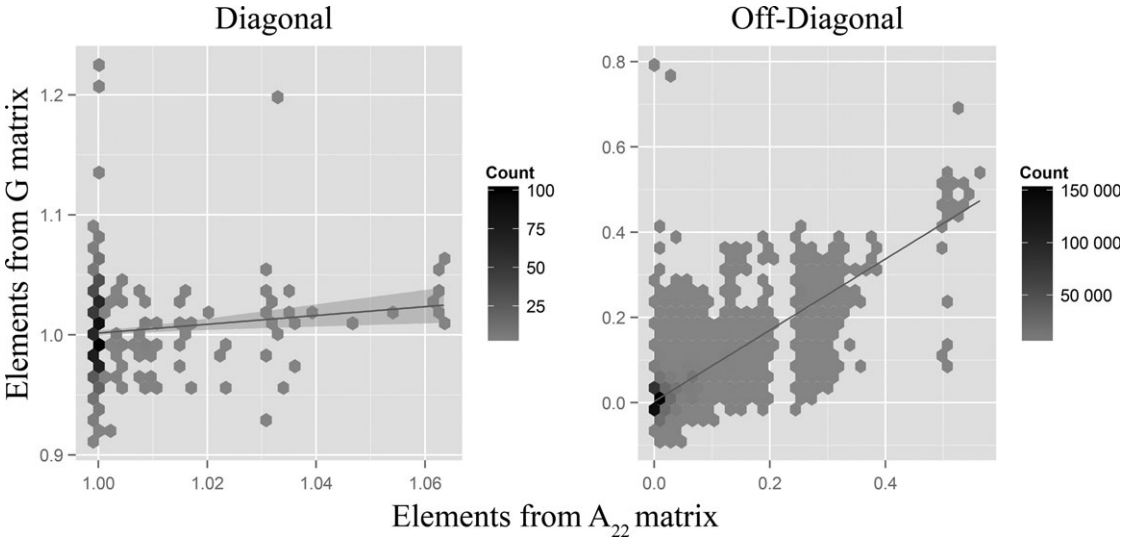

Figure 2 Elements of diagonal and off-diagonal of $\mathbf{A_{22}}$ (sub-matrix of ***A*** for genotyped animals) and ***G*** (genomic matrix) matrices plotted against each other (the grey line is a regression line and the shadow area represented the 95% confidence intervals).

from first to ninth parity and with days in milk between six and 365. In general, few studies have given the heritability of LP in bovine milk despite its importance as the major carbohydrate and osmolyte of milk, determining milk volume, and as raw material in the manufacture of dairy products such as cheese and whey. Also, a favourable effect of lactose content on clotting time was reported in an Ayrshire population (Lindström *et al.* 1984). Nevertheless, because of its moderate to high heritability and positive genetic correlation with PP and FP, which are common selection criteria in animal breeding; indirect selection for LP is likely to occur.

Low heritabilities for daily MY also were estimated by Cassandro *et al.* (2008) and Penasa *et al.* (2015), with values of 0.09 and 0.104, respectively; and for SCS by Cassandro *et al.* (2008) and Negussie *et al.* (2008), with values ranging from 0.07 and 0.12. However, differences among studies regarding heritability estimates for SCS were expected due to the variety of equations that can be used to transform somatic cell count to somatic cell score. Herein, the estimates for SCS presented large standard errors, even with the inclusion of genotypic data, probably because of the high variability of this trait. This way, a greater amount of information is necessary to obtain more accurate estimates of genetic parameters for SCS.

Differences in the analysis model and in the unit and methodology of measurement can make it difficult to compare genetic parameters for fatty acids content among studies (Bastin *et al.* 2011). For example, in Soyeurt *et al.* (2008) and Bastin *et al.* (2013), calibration equations were used to predict fatty acids composition in milk by mid-infrared spectrometry. These equations were built by partial least squares regressions from chromatographic and spectral data (Soyeurt *et al.* 2006, 2011). In turn, in our study, the measurements obtained by mid-infrared spectrometry technique were used directly, with a previous validation analysis showing correlations ranging from 0.60 to 0.92 between the measurements determined by gas chromatography and mid-infrared spectrometry (Rodriguez *et al.* 2014). Moreover, these traits are greatly influenced by environmental conditions, mainly by the diet (Penasa *et al.* 2015). Indeed, higher heritabilities were estimated by Soyeurt *et al.* (2008) for SFA ($h^2 = 0.42$) and MUFA ($h^2 = 0.14$), and by Bastin *et al.* (2011), with values of 0.426 for SFA, 0.212 for MUFA, 0.298 for PUFA, 0.223 for UFA, 0.408 for C16:0, 0.380 for C18:0, and 0.179 for C18:1. Nevertheless, similar estimates were obtained by Penasa *et al.* (2015), with values of 0.246, 0.069, 0.082 and 0.078 for SFA, UFA, MUFA and PUFA, respectively. Herein and in these studies, the heritabilities associated with saturated fatty acids (SFA, C18:0, and C16:0) were higher than those obtained for unsaturated fatty acids (UFA, MUFA, PUFA, and C18:1). This probably occurs because of the origin of fatty acids. Short and medium-chain saturated fatty acids are mainly synthetized *de novo* in the mammary gland through the action of acetyl-coenzyme A carboxylase and fatty acid synthase whereas a fraction of palmitic acid and other long chain fatty acids are obtained from the blood stream, derived from the diet

or body fat mobilization (Grummer 1991; Lock & Bauman 2004; Bastin *et al.* 2011). Thus, due to the greater environmental influence over long chain fatty acids, the lower heritability associated with this group is reasonable (Bastin *et al.* 2011).

Despite the possibility of changing milk fatty acids profile through sire selection, the direction of this selection is not well established yet, mainly because of the variety of aspects linked to these traits. Among saturated fatty acids, the butyric acid is a modulator of gene function, acting in colon cancer protection and inhibiting mammary tumorigenesis (Parodi 1997; German 1999; Haug *et al.* 2007); the caprylic and capric acids have possible antiviral activities (Thormar *et al.* 1994) whereas lauric, myristic and palmitic acids can increase levels of low (LDL) and high (HDL) density lipoprotein cholesterol (Haug *et al.* 2007); although no consistent association between dairy foods and cardiovascular diseases has been reported (Haug *et al.* 2007; German *et al.* 2009).

At the same time, monounsaturated fatty acids, such as oleic acid, were related to lower plasma cholesterol, LDL-cholesterol and triacylglycerol concentrations (Kris-Etherton *et al.* 1999). Omega-3 fatty acids (docosahexaenoic acid and eicosapentaenoic acid) can act in heart diseases prevention since they have anti-inflammatory effects, are antithrombotic and inhibit atherosclerosis, among other properties (Connor 2000). Another group of PUFA, conjugated linoleic acid (CLA) was associated with antiadipogenic, antidiabetogenic, anticarcinogenic and antiatherosclerotic effects (Belury 2002; Lock & Bauman 2004). Nevertheless, an increase in UFA content can change the sensory quality of dairy products (Chilliard & Ferlay 2004; Woods & Fearon 2009), increasing their susceptibility to oxidation and development of off-flavours (Ashes *et al.* 1997; Campbell *et al.* 2003). Moreover, high levels of long-chain fatty acids in milk fat, especially stearic and oleic acids; may be due to body fat mobilization in subclinical ketotic cows, indicating a negative energy balance (Van Haelst *et al.* 2008).

Considering these properties, several studies recommended an optimum proportion of fatty acids in milk to be achieved through sire selection (Grummer 1991; Chilliard *et al.* 2000; Haug *et al.* 2007). However, since they are highly correlated with fat percentage, indirect selection is already happening. Therefore, it is important to rapidly establish the priority aspects related to fatty acids profile (human health, cow health, manufacturing features) (Bastin *et al.* 2013) and also to evaluate the possible effects of selection for some fatty acids over fatty acids profile and other milk composition traits, such as protein and lactose.

The moderate to high heritabilities associated with milk components, especially FP and PP, suggest that sire selection can be used to improve milk quality. However, the antagonist relationship between MY and these milk components, also reported by Miglior *et al.* (2007), Cassandro *et al.* (2008) and Penasa *et al.* (2015), can prevent a simultaneous genetic gain for milk volume and milk quality. This is particularly harmful when the payment for the farmer is predominantly based on the amount of milk delivered, making milk yield the main selection objective as occurs in Brazil. Such a situation confirms the usefulness of a total merit index in selection.

Genetic correlations between SCS and milk yield and composition traits varied among studies, mainly because of the already reported effects of stage of lactation and parity (Haile-Mariam *et al.* 2001). The estimates obtained in the present study should be interpreted with caution due to their high standard errors. Herein, the negative genetic correlation between MY and SCS was expected because the occurrence of mastitis, indicated by the high SCS, damages the udder with a consequent reduction in milk yield. However, a positive correlation (unfavourable) can also occur, given that cows with high production may be more susceptible to infection (Haile-Mariam *et al.* 2001). The harm caused by mastitis in the udder can also explain the small decrease in protein and fat content with even greater changes in the individual milk components. The concentration of proteins synthesized in the mammary gland in milk, such as α-casein, β-casein and β-lactoglobulin, are often reduced in the presence of mastitis concomitantly with an increase of bovine serum albumin and immunoglobulins (Kitchen 1981). Similarly, Randolph & Erwin (1974) reported higher concentrations of short-chain esterified fatty acids and unsaturated fatty acids and lower concentration of esterified fatty acids in mastitis-positive milks compared to mastitis-negative milks. The possible causes are alterations in direct synthesis of lipid components by the mammary gland or to changes in permeability of the mammary gland (Randolph & Erwin 1974). This behaviour was confirmed in the present study, with positive genetic correlations between SCS and UFA, MUFA, PUFA and C18:1, and negative genetic correlations between SCS and SFA, C16:0 and C18:0.

Somatic cell score also has a negative genetic correlation with LP. This antagonist relationship can be due to tissue damage caused by the infection and the consequent decrease of lactose biosynthesis, as explained for PP and FP above. Another hypothesis is a reduction of available glucose to the mammary

gland as a result of reduced blood flow caused by the general stress conditions during the disease (Kitchen 1981).

The differences among studies were attributed to possible effects of lactation number, days in milk and methodology adopted. However, these differences also could be caused by the production environment. Costa *et al.* (2000), using fat yield and milk yield data of daughters of 705 United States sires and 701 Brazilian sires, estimated lower heritabilities in the Brazilian population, with values of 0.25 and 0.22 for milk yield and fat yield, respectively, in comparison to 0.34 and 0.35 obtained for the same traits in the US population. This probably occurs because environmental conditions limited the expression of genetic potential, constraining the genetic variance and, consequently, reducing heritability. Some implications are derived from this situation. Firstly, it is more difficult to differentiate among breeding values, which will demand greater selection intensity (Costa *et al.* 2000) and more accurate predictions to achieve genetic progress through selection. Thus, even though the inclusion of genomic information did not alter the estimates of genetic parameters, genotype records can be useful in this situation to increase the amount of data available per individual, enhancing the prediction of genetic merit. Secondly, the relative superiority of sires selected in other countries may be less strongly related to breeding values under tropical and subtropical conditions with a consequent lower response to selection (Costa *et al.* 2000). These results confirm the necessity to locally estimate the genetic parameters and, furthermore, the importance to evaluate the animals based on the conditions where selection decisions will be made.

Conclusions

Smaller estimates of components of covariance were obtained here compared to other similar studies. These differences can be due to lactation number, days in milk, methodology used or even to management and environment factors, in this latter case indicating that tropical and subtropical conditions can restrict the expression of genetic potential by individuals with a consequent reduction of genetic variability. In such a situation, the differentiation among animals is problematic, highlighting the importance of using parameters specific of the population in which selection will be applied. Even though genomic information had little impact on estimates of genetic parameters, it can be an auxiliary tool to accurately predicted genetic merit and, therefore, increase genetic gain when the superiority of selected parents is less evident. The greatest impact of genomics is in reducing the generation interval with only a small reduction in accuracy leading to a large increase in rate of genetic progress.

The moderate and high heritabilities for milk components revealed the possibility of improving milk quality by using selection. Nevertheless, because of the antagonist relationship between milk yield and milk component traits and also, the high and unfavourable correlation between milk fat content and total saturated fatty acids in milk, it should be valuable to consider those traits simultaneously in genetic evaluation, considering their economic relevance for the system and their phenotypic and genetic correlation.

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References

Aguilar I., Misztal I., Johnson D.L., Legarra A., Tsuruta S., Lawlor T.J. (2010) A unified approach to utilize phenotypic, full pedigree, and genomic information for genetic evaluation of Holstein final score. *J. Dairy Sci.*, **93**, 743–752.

Aka A., Shook G.E. (1980) An optimum transformation for somatic cell concentration in milk. *J. Anim. Sci.*, **63**, 487–490.

Ashes J.R., Gulati S.K., Scott T.W. (1997) Potential to alter the content and composition of milk fat through nutrition. *J. Dairy Sci.*, **80**, 2204–2212.

Bastin C., Gengler N., Soyeurt H. (2011) Phenotypic and genetic variability of production traits and milk fatty acid contents across days in milk for Wallloon Holstein first-parity cows. *J. Dairy Sci.*, **94**, 4152–4163.

Bastin C., Soyeurt H., Gengler N. (2013) Genetic parameters of milk production traits and fatty acid contents in milk for Holstein cows in parity 1–3. *J. Anim. Breed. Genet.*, **130**, 118–127.

Belury M.A. (2002) Dietary conjugated linoleic acid in health: physiological effects and mechanisms of action. *Annu. Rev. Nutr.*, **22**, 505–531.

Campbell W., Drake M.A., Larick D.K. (2003) The impact of fortification with conjugated linoleic acid (CLA) on the quality of fluid milk. *J. Dairy Sci.*, **86**, 43–51.

Cassandro M., Comin A., Ojala M., Dal Zotto R., De Marchi M., Gallo L., Carnier P., Bittante G. (2008) Genetic parameters of milk coagulation properties and their relationships with milk yield and quality traits in Italian Holstein cows. *J. Dairy Sci.*, **91**, 371–376.

Chilliard Y., Ferlay A. (2004) Dietary lipids and forages interactions on cow and goat milk fatty acid composition and sensory properties. *Reprod. Nutr. Dev.*, **44**, 467–492.

Chilliard Y., Ferlay A., Mansbridge R., Doreau M. (2000) Ruminant milk fat plasticity: nutritional control of saturated, polyunsaturated, trans and conjugated fatty acids. *Ann. Zootechnol.*, **49**, 181–205.

Christensen O.F., Madsen P., Nielsen B., Ostersen T., Su G. (2012) Single-step methods for genomic evaluation in pigs. *Animal*, **10**, 1565–1571.

Cienfuegos-Rivas E.G., Oltenacu P.A., Blake R.W., Schwager S.J., Castillo-Juarez H., Ruiz F.J. (1999) Interaction between milk yield of Holstein cows in Mexico and the United States. *J. Dairy Sci.*, **82**, 2218–2223.

Connor W.E. (2000) Importance of n-3 fatty acids in health and disease. *Am. J. Clin. Nutr.*, **71**, 171S–175S.

Costa C.N., Blake R.W., Pollak E.J., Oltenacu P.A., Quaas R.L., Searle S.R. (2000) Genetic analysis of Holstein cattle populations in Brazil and the United States. *J. Dairy Sci.*, **83**, 2963–2974.

Curtis S.E. (1983) Environmental Management in Animal Agriculture, 1st edn. The Iowa State Universtity Press, Ames, IA, USA.

Ding X., Zhang Z., Li X., Wang S., Wu X., Sun D., Yu Y., Liu J., Wang Y., Zhang Y., Zhang S., Zhang Y., Zhang Q. (2013) Accuracy of genomic prediction for milk production traits in the Chinese Holstein population using a reference population consisting of cows. *J. Dairy Sci.*, **96**, 5315–5323.

Enjalbert F., Nicot M.C., Bayourthe C., Moncoulon R. (1998) Duodenal infusions of palmitic, stearic or oleic acids differently affect mammary gland metabolism of fatty acids in lactating dairy cows. *J. Nutr.*, **128**, 1525–1532.

German J.B. (1999) Butyric acid: a role in cancer prevention. *Nutr. Bull.*, **24**, 203–209.

German J.B., Gibson R.A., Krauss R.M., Nestel P., Lamarche B., van Staveren W.A., Steijns J.M., de Groot L.C.P.G.M., Lock A.L., Destaillats F. (2009) A reappraisal of the impact of dairy foods and milk fat on cardiovascular disease risk. *Eur. J. Nutr.*, **48**, 191–203.

Grummer R.R. (1991) Effect of feed on the composition of milk fat. *J. Dairy Sci.*, **74**, 3244–3257.

Haile-Mariam M., Bowman P.J., Goddard M.E. (2001) Genetic and environmental correlations between test-day somatic cell count and milk yield traits. *Liv. Prod. Sci.*, **73**, 1–13.

Haile-Mariam M., Nieuwhof G.J., Beard K.T., Konstatinov K.V., Hayes B.J. (2013) Comparison of heritabilities of dairy traits in Australian Holstein-Friesian cattle from genomic and pedigree data and implications for genomic evaluations. *J. Anim. Breed. Genet.*, **130**, 20–31.

Haug A., Hostmark A.T., Harstad O.M. (2007) Bovine milk in human nutrition – a review. *Lipids Health Dis.*, **6**, 25.

Kitchen B.J. (1981) Review of the progress of Dairy Science: Bovine mastitis: milk compositional changes and related diagnostic tests. *J. Dairy Res.*, **48**, 167–188.

Koivula M., Standén I., Su G., Mäntysaari E.A. (2012) Different methods to calculate genomic predictions – comparisons of BLUP at the single nucleotide polymorphism level (SNP-BLUP), BLUP at the individual level (G-BLUP), and the one-step approach (H-BLUP). *J. Dairy Sci.*, **95**, 4065–4073.

Krag K., Poulsen N.A., Larsen M.K., Larsen L.B., Janss L.L., Buitenhuis B. (2013) Genetic parameters for milk fatty acids in Danish Holstein cattle based on SNP markers using a Bayesian approach. *BMC Genet.*, **14**, 79.

Kris-Etherton P.M., Pearson T.A., Wan Y., Hargrove R.B., Moriarty K., Fishell V., Etherton T.D. (1999) High-monounsaturated fatty acid diets lower both plasma cholesterol and triacylglycerol concentrations. *Am. J. Clin. Nutr.*, **70**, 1009–1015.

Lindström U.B., Antila V., Syväjärvi J. (1984) A note on some genetic and non-genetic factors affecting clotting time of Ayrshire milk. *Acta Agric. Scand.*, **34**, 349–355.

Lock A.L., Bauman D.E. (2004) Modifying milk fat composition of dairy cows to enhance fatty acids beneficial to human health. *Lipids*, **39**, 1197–1206.

McDonald J.S. (1968) Radiographic method for anatomic study of the teat canal. Changes with lactation age. *Am. J. Vet. Res.*, **29**, 1207–1210.

Mensink R.P., Zock P.L., Kester A.D.M., Katan M.B. (2003) Effects of dietary fatty acids and carbohydrates on the ration of serum total to HDL cholesterol and on serum lipids and apolipoproteins: a meta-analysis of 60 controlled trials. *Am. J. Clin. Nutr.*, **77**, 1146–1155.

Meyer K., Houle D. (2013) Sampling based approximation of confidence intervals for functions of genetic covariance matrices. In: Proceedings of the 20th Association for the Advancement of Animal Breeding and Genetics Conference. Napier (New Zeland), 523–526 October 2013.

Miglior F., Sewalem A., Jamrozik J., Bohmanova J., Lefebvre D.M., Moore R.K. (2007) Genetic analysis of milk urea nitrogen and lactose and their relationships with other production traits in Canadian Holstein cattle. *J. Dairy Sci.*, **90**, 2468–2479.

Misztal I., Tsuruta S., Strabel T., Auvray B, Druet T., Lee D. (2002) BLUPF90 and related programs (BGF90). In: Proceedings of the 7th World Congress on Genetics Applied

to Livestock Production, Montpellier (France). CD-ROM Communication.
Misztal I., Aggrey S.E., Muir W.M. (2013) Experiences with a single-step genome evaluation. *Poultry Sci.*, **92**, 2530–2534.
Negussie E., Strandén I., Mäntysaari E.A. (2008) Genetic association of clinical mastitis with test-day somatic cell score and milk yield during first lactation of Finnish Ayrshire cows. *J. Dairy Sci.*, **91**, 1189–1197.
Parodi P.W. (1997) Cows' milk fat components as potential anticarcinogenic agents. *J. Nutr.*, **127**, 1055–1060.
Patry C., Ducrocq V. (2011) Evidence of biases in genetic evaluations due to genomic preselection in dairy cattle. *J. Anim. Sci.*, **94**, 1011–1020.
Penasa M., Tiezzi F., Gottardo P., Cassandro M., De Marchi M. (2015) Genetics of milk fatty acid groups predicted during routine data recording in Holstein dairy cattle. *Livest. Sci.*, **173**, 9–13.
Randolph H.E., Erwin R.E. (1974) Influence of mastitis on properties of milk. X. Fatty acid composition. *J. Dairy Sci.*, **57**, 865–868.
Rodriguez M.A.P., Petrini J., Ferreira E.M., Mourão L.R.M.B., Salvian M., Cassoli L.D., Pires A.V., Machado P.F., Mourão G.B. (2014) Concordance analysis between estimation methods of milk fatty acids content. *Food Chem.*, **156**, 170–175.
Schopen G.C.B., Heck J.M.L., Bovenhuis H., Visker M.H.P.W., van Valenberg H.J.F. (2009) Genetic parameters for major milk proteins in Dutch Holstein-Friesians. *J. Dairy Sci.*, **92**, 1182–1191.
Soyeurt H., Dardenne P., Dehareng F., Lognay G., Veselko D., Marlier M., Bertozzi C., Mayeres P., Gengler N. (2006) Estimating fatty acid content in cow milk using mid-infrared spectrometry. *J. Dairy Sci.*, **89**, 3690–3695.
Soyeurt H., Dardenne P., Dehareng F., Bastin C., Gengler N. (2008) Genetic parameters of saturated and monounsaturated fatty acid content and the ratio of saturated to unsaturated fatty acids in bovine milk. *J. Dairy Sci.*, **91**, 3611–3626.
Soyeurt H., Dehareng F., Gengler N., McParland S., Wall E., Berry D.P., Coffey M., Dardenne P. (2011) Mid-infrared prediction of bovine milk fatty acids across multiple breeds, production systems, and countries. *J. Dairy Sci.*, **94**, 1657–1667.
Stoop W.M., van Arendonk J.A.M., Heck J.M.L., van Valenberg H.J.F., Bovenhuis H. (2008) Genetic parameters for major fatty acids and milk production traits of Dutch Holstein-Friesians. *J. Dairy Sci.*, **91**, 385–394.
Thormar H., Isaacs E.E., Kim K.S., Brown H.R. (1994) Interaction of visna virus and other enveloped viruses by free fatty acids and monoglycerides. *Ann. N. Y. Acad. Sci.*, **724**, 465–471.
Tiezzi F., Pretto D., De Marchi M., Penasa M., Cassandro M. (2013) Heritability and repeatability of milk coagulation properties predicted by mid-infrared spectroscopy during routine data recording, and their relationships with milk yield and quality traits. *Animal*, **10**, 1592–1599.
Tsuruta S., Misztal I., Lawlor T.J. (2013) Genomic evaluations of final score for US Holsteins benefit from the inclusion of genotypes on cows. *J. Dairy Sci.*, **96**, 3332–3335.
Van Haelst Y.N.T., Beeckman A., Van Knegsel A.T.M., Fievez V. (2008) Elevated concentrations of oleic acid and long-chain fatty acids in milk fat of multiparous subclinical ketotic cows. *J. Dairy Sci.*, **91**, 4683–4686.
VanRaden P.M. (2008) Efficient methods to compute genomic predictions. *J. Dairy Sci.*, **91**, 4414–4423.
VanRaden P.M. (2015) findhap.f90 – Find haplotypes and impute genotypes using multiple chip sets and sequence data (available at: http://aipl.arsusda.gov/software/findhap/; last accessed 20 April 2015).
Veerkamp R.F., Mulder H.A., Thompson R., Calus M.P.L. (2011) Genomic and pedigree-based genetic parameters for scarcely recorded traits when some animals are genotyped. *J. Dairy Sci.*, **94**, 4189–4197.
Walsh S., Buckley F., Berry D.P., Rath M., Pierce K., Byrne N., Dillon P. (2007) Effects of breed, feeding system, and parity on udder health and milking characteristics. *J. Dairy Sci.*, **90**, 5767–5779.
West J.W. (2003) Effects of heat-stress on production in dairy cattle. *J. Dairy Sci.*, **86**, 2131–2144.
Woods V.B., Fearon A.M. (2009) Dietary sources of unsaturated fatty acids for animals and their transfer into meat, milk, and eggs: a review. *Livest. Sci.*, **126**, 1–20.