Livestock Science 180 (2015) 58–64

Contents lists available at ScienceDirect

Livestock Science

journal homepage: www.elsevier.com/locate/livsci

Heritability of milk fat composition is considerably lower for Meuse-Rhine-Yssel compared to Holstein Friesian cattle

M.H.T. Maurice-Van Eijndhoven [a,b], R.F. Veerkamp [a], H. Soyeurt [c,d], M.P.L. Calus [a,*]

[a] Animal Breeding and Genomics Centre, Wageningen UR Livestock Research, P.O. Box 338, 6700 AH Wageningen, The Netherlands
[b] Animal Breeding and Genomics Centre, Wageningen University, P.O. Box 338, 6700 AH Wageningen, The Netherlands
[c] Agriculture, Bio-engineering and Chemistry Department, Gembloux Agro-Bio Tech, University of Liège, 5030 Gembloux, Belgium
[d] National Fund for Scientific Research, 1000 Brussels, Belgium

ARTICLE INFO

Article history:
Received 29 September 2014
Received in revised form
25 May 2015
Accepted 13 July 2015

Keywords:
Fatty acids
Infrared spectrometry
Genetic variability
Cattle breeds

ABSTRACT

The aim of this paper is to identify differences in genetic variation of fatty acid (FA) composition in milk in different breeds. Data used included Meuse-Rhine-Yssel (MRY) and Holstein Friesian (HF) cattle breeds which were raised in the Netherlands. Both populations participated in the same milk recording system, but differed in selection history, where in the MRY there has been relatively very little emphasis on selection for high-input high-output production systems compared to HF. Differences in genetic variation were investigated by estimating breed specific additive genetic variances and heritabilities for FA contents in milk of MRY and HF. Mid Infrared Spectrometry spectra were used to predict total fat percentage and detailed FA contents in milk (14 individual FA and 14 groups of FA in g of fat/dL of milk). The dataset for MRY contained 2916 records from 2049 registered cows having at least 50% genes of MRY origin and the dataset used for HF contained 155,319 records from 96,315 registered cows having at least 50% genes of HF origin. Variance components of individual FA content in milk for the different breeds were estimated using a single trait animal model. Additive genetic variances for FA produced through de novo synthesis (short chain FA, C12:0, C14:0, and partly C16:0), C14:1 *c*-9 and C16:1 *c*-9 were significantly higher ($P < 0.001$) for HF compared to MRY. Heritabilities of the individual FA, C4:0 to C18:0, for HF ranged from 0.28 to 0.52 and for MRY from 0.17 to 0.34. Heritabilities of the individual C18 unsaturated FA for HF ranged from 0.11 to 0.34 and for MRY from 0.10 to 0.26. Although the mean content in milk for the FA C18:2 *c*-9, *t*-11 was low in both breeds, the additive genetic variance in our dataset was significantly higher for MRY ($P < 0.05$) compared to HF. Heritabilities of the groups of FA for HF ranged from 0.19 to 0.53 and for MRY from 0.11 to 0.28. For the majority of the FA, the additive genetic variances for HF were significantly higher compared to MRY, except for most of the poly-unsaturated FA. The results for the poly-unsaturated FA, however, may be affected by the lower accuracy of the predictions for these FA. In conclusion, our results show that the HF breed has substantially larger genetic variance for most FA compared to MRY.

1. Introduction

Fat is one of the major components in bovine milk. Bovine milk fat is composed of a wide range of fatty acids (FA) which content and composition in the milk vary between cows. Extending the knowledge on variation in detailed milk fat composition among cows is of interest for the dairy industry because fat composition is associated with processability (e.g. Smet et al., 2009), human health (e.g. Mensink et al., 2003; Palmquist et al., 2006) and also methane emission (Dijkstra et al., 2011). The variation in FA composition in milk between cows is partly due to environmental effects, mainly differences in cows diet (e.g. Baumgard et al., 2001; Sterk et al., 2011), lactation stage (e.g. Stull et al., 1966), and also a considerable part of the variation has a genetic origin within and across lactation (e.g. Stoop et al., 2008; Bastin et al. 2011). Considering the heritabilities reported by Stoop et al. (2008), ranging from 0.22 to 0.71, there are possibilities for the dairy industry to modify FA composition in milk of Dutch HF cows using breeding strategies. An important unanswered question is whether the same applies for other Dutch local breeds like the Meuse-Rhine-Yssel (MRY), in which there has been relatively very little emphasis on selection for high-input high output production systems used in the dairy sector. Differences for the FA profile of milk fat between breeds have been described by e.g. Maurice-Van

* Corresponding author. Fax: +31 320 293 591.
E-mail address: mario.calus@wur.nl (M.P.L. Calus).

http://dx.doi.org/10.1016/j.livsci.2015.07.009
1871-1413/

Eijndhoven et al. (2013) reporting higher content of saturated FA (SAT) produced by Jersey cows compared to a number of local Dutch breeds. Although Maurice-Van Eijndhoven et al. (2013) reported no differences in the level of FA composition between the Dutch MRY and HF, differences in within breed variability for both breeds need to be known to assess whether alternative breeds like MRY can contribute to breeding strategies to change FA composition. The aim of this paper was to identify differences in genetic variation within the MRY and HF cattle breeds, based on predicted FA composition in milk, using Mid-Infrared Spectrometry (MIRS) spectra on a large dataset. This was achieved by estimating breed specific additive genetic variances and heritabilities for FA composition of MRY and HF.

2. Materials and methods

The MRY and HF populations used in the analysis, both participated in the same milk recording system, but differed in selection history, where in the MRY there has been relatively very little emphasis on selection for high-input high-output production systems compared to HF. The numbers of MRY cows in the Netherlands participating in milk recording have decreased from around 100,000 in 1980 to 5500 in 2014, while in the same period the number of HF cows has changed from 4000 to more than 400,000 (CRV BV, Arnhem, The Netherlands).

2.1. Data collection and data editing

MIRS spectra of milk samples were collected via the Dutch milk recording system of CRV BV (Arnhem, The Netherlands) between October and December 2006. Samples were treated immediately with 0.03% (wt/wt) sodium azide to avoid microbiological growth. The MIRS spectra were obtained using 3 Fourier-transformed interferogram machines (MilkoScan FT 6000, Foss Electric, Denmark) at the laboratory of Qlip N.V. (Leusden, The Netherlands). The 1886 sampled herds were a random representation of all herds participating in the milk recording system of CRV BV.

The initial dataset contained 372,429 test-day records of 230,995 cows. Data-editing steps included the deletion of records and cows for the following reasons: less than 75% of the breed composition known, unknown sire, incomplete milk recording data (e.g. unknown birthdate or DIM), multiple records from the same cow on the same sample date, cows with records in multiple herds, cows reported sick at sampling date, cows in parity 11 or higher, cows before 5 or after 365 days in lactation, and records from herds with less than 5 purebred cows of the same breed (HF, MRY, Dutch Friesian (DF), or Groningen White Headed (GWH)) per herd. To detect records with possible errors, due to, for example, swapped samples, fat content recorded via the regular milk control (predicted by QLIP N.V.) was compared to fat content obtained using the RobustMilk prediction equations that were developed by Soyeurt et al. (2011). The correlation coefficient between fat content predicted by QLIP N.V. and fat content predicted using the RobustMilk prediction equations was 0.996. When the absolute difference in both predictions for fat percentage was more than 0.35 the record was removed. Finally, complete records with extreme outliers in at least 1 of all predicted traits (+/− 5 SD of the mean) were deleted. After these editing steps the dataset contained 307,656 records. Because of computational limitations, this dataset was reduced by randomly eliminating ~50% of the herds with only HF cows (at least 75% HF, i.e. herds without any pure- or crossbred MRY, DF, or GWH cows). The dataset used for MRY contained 2916 records of in total 2049 cows registered having at least 50% genes of MRY origin with a pedigree of 13,506 animals and the dataset used to estimate heritabilities for HF contained 155,319 records of in total 96,315 cows registered having at least 50% genes of HF origin with a pedigree of 405,968 animals. Pedigree files for both data sets included all known ancestors as far back as possible. Ancestors with unknown parents and only 1 offspring in the pedigree were removed.

2.2. Measuring fatty acid composition

Detailed milk composition on milk basis (g of FA/dL milk) of the 14 individual FA (C4:0, C6:0, C8:0, C10:0, C12:0, C14:0, C14:1 *c*-9, C16:0, C16:1 *c*-9, C18:0, C18:1 *c*-9, C18:2 *c*-9, 12, C18:3 *c*-9, 12, 15, and C18:2 *c*-9, *t*-11) and the 14 groups of FA [total trans C18:1, total cis C18:1, total C18:2, total trans C18, total SFA, total mono-unsaturated FA (MUFA), total poly-unsaturated FA (PUFA), total UFA, short-chain FA (SCFA), medium-chain FA (MCFA), long-chain FA (LCFA), total *n*-3 FA, total *n*-6 FA, and total branched-chain FA (BCFA)] were predicted from the MIRS spectra. For those predictions, updated versions of the RobustMilk calibration equations published by Soyeurt et al. (2011) were used, that were based on 1236 milk samples from multiple breeds and countries (calibration equations were updated by expanding the number of samples used in the calibration data set from 570 to 1236). The method used to relate MIRS spectra to FA data was partial least square regression after a first derivative pre-treatment on spectral data to correct the baseline drift. A T-outlier test was also used during the calibration process to delete potential outliers based on the gas chromatographic measurements. Therefore the final number of samples included in each calibration equation varied following the considered FA. More detailed information about the methodology used to develop the calibration equations is given by Soyeurt et al. (2011). The definition of the groups of FA are given in Table 1 and some descriptive statistics of the calibration equations, which are described by Soyeurt et al. (2011), are given in Table 2. More detailed descriptive statistics of the calibration equations are published by Maurice-Van Eijndhoven et al. (2012) including an external validation for the MRY breed.

Next to the detailed FA composition, 5 production traits were analysed (milk yield, fat%, protein%, fat yield and protein yield). Fat content was predicted using the RobustMilk calibration equations published by Soyeurt et al. (2011) and protein content was predicted by QLIP N.V.

2.3. Statistical analysis

Genetic variances were estimated in separate analyses for HF and MRY in ASReml 3.0 (Gilmour et al., 2009) using the following animal model:

$$y_{ijklmnopqrstuv} = \mu + b_1 \times \mathrm{DIM}_i + b_2 \times \exp^{-0.05\times \mathrm{dim}}{}_i + \mathrm{parity}_j + b_3 \times \mathrm{age}_k\left(\mathrm{parity}_j\right) + \mathrm{htd}_l + b_4 \times \mathrm{HF}_m + b_5 \times \mathrm{MRY}_n + b_6 \times \mathrm{DF}_o + b_7 \times \mathrm{GWH}_p + b_8 \times \mathrm{JER}_q + b_9 \times \mathrm{HET}_r + b_{10} \times \mathrm{REC}_s + a_t + \mathrm{pe}_t + e_{ijklmnopqrstu},$$

where $y_{ijklmnopqrstuv}$ was the dependant variable for cow t in days in milk (DIM) i, with parity j, calving age k, producing at herd test date (htd) l, and having a breed composition $mnopq$ for HF (m), MRY (n), DF (o), GWH (p), and JER (q). The μ was the overall mean of the model; b_1 was the fixed regression coefficient on DIM_i and b_2 was the fixed regression coefficient on DIM_i modelled with a Wilmink curve (Wilmink, 1987); parity_j was a fixed effect with 4 classes for corresponding lactation numbers of parity 1, 2, and 3 and the 4th class included parity 4–10; b_3 was the fixed regression coefficient on age_k, which was calving age in days, within the jth parity; htd_l was a fixed effect defining groups of

Table 1
Definition of the groups of fatty acids.

Group	Fatty acids
Total *t* C18:1	C18:1 *t*-6-11; C18:1 *t*-12-14
Total *c* C18:1	C18:1 *c*-9; C18:1 *c*-11; C18:1 *c*-12; C18:1 *c*-13; C18:1 *c*-14; C18:1 *t*-16
Total C18:2	C18:2 ∑ ttNMID; C18:2 *c*-9, *t*-13; C18:2 *t*-8, *c*-12; C18:2 *c*-9, *t*-12; C18:2 *t*-8, *c*-13; C18:2 *t*-11, *c*-15; C18:2 *t*-9, *c*-12; C18:2 *c*-9, *c*-12
Total *t* C18	C18:1 *t*6-11; C18:1 *t*12-14; C18:2 ∑ ttNMID; C18:2 *c*-9, *t*-13; C18:2 *t*-8, *c*-12; C18:2 *c*-9, *t*-12; C18:2 *t*-8, *c*-13; C18:2 *t*-11, *c*-15; C18:2 *t*-9, *c*-12
Saturated fatty acids (SFA)	C4:0; C6:0; C8:0; C10:0; C12:0; C13:0 *iso*; C13:0 *ante-iso*; C14:0; C14:0 *iso*; C15:0; C15:0 *iso*; C15:0 *ante-iso*; C16:0; C16:0 *iso*; C17:0; C17:0 *iso*; C17:0 *ante-iso*; C18:0; C18:0 *iso*; C19:0; C20:0; C22:0
Unsaturated fatty acids (UFA)	MUFA; PUFA
Unsaturated fatty acids with 1 double bound (MUFA)	C10:1; C12:1 *cis*; C14:1 *cis*; C16:1 cis; C16:1 *trans*; C17:1; C18:1 *c*-9; C18:1 *c*-11; C18:1 *c*-12; C18:1 *t*-6-11; C18:1 *t*-12-14; C18:1 *c*-13; C18:1 *c*-14;C18:1 *t*-16; C20:1 *c*-9; C20:1*c*-11
Unsaturated fatty acids with 2 or more double bounds (PUFA)	C18:2 ∑ ttNMID; C18:2 *c*-9, *t*-13; C18:2 *t*-8, *c*-12; C18:2 *c*-9, *t*-12; C18:2 *t*-8, *c*-13; C18:2 *t*-11, *c*-15; C18:2 *t*-9, *c*-12; C18:2 *c*-9,*c*-12; C18:3 *c*-9, c-12, *c*-15; C18:2 *c*-9, *t*-11 (CLA); C20:3 ($n-6$); C20:4 ($n-6$); C20:5 EPA ($n-3$); C22:5 DPA
Short chain fatty acids (SCFA)	C4–C10
Medium chain fatty acids (MCFA)	C12–C16
Long chain fatty acids (LCFA)	C17–C22
$n-3$	C18:3 *c*-9,c-12, *c*-15; C20:5 (EPA); C22:5 (DPA)
$n-6$	C18:2 ∑ ttNMID; C18:2 *c*-9, *t*-13; C18:2 *t*-8, *c*-12; C18:2 *c*-9, *t*-12; C18:2 *t*-8, *c*-13; C18:2 *t*-11, *c*-15; C18:2 *t*-9, *c*-12; C18:2 *c*-9, *c*-12; C20:3 ($n-6$); C20:4 ($n-6$)
BCFA	C13:0 *iso*; C13:0 *ante-iso*; C14:0 *iso*; C15:0 *iso*; C15:0 *ante-iso*; C16:0 *iso*; C17:0 *iso*; C17:0 *ante-iso*; C18:0 *iso*

Table 2
Descriptive statistics of the fatty acid calibration equations and data used to derive the equations.

Traits	N[a]	R^2cv[b]	RPD[c]
Production traits			
Fat%	1166	1.00	33.53
Traits g/dL milk			
C4:0	1186	0.93	3.68
C6:0	1189	0.96	4.81
C8:0	1180	0.96	5.00
C10:0	1183	0.96	4.72
C12:0	1180	0.95	4.61
C14:0	1184	0.95	4.70
C14:1 *c*-9	1180	0.78	2.13
C16:0	1179	0.97	6.20
C16:1 *c*-9	1179	0.78	2.14
C18:0	1173	0.90	3.24
C18:1 *c*-9	1194	0.96	5.06
Total cis C18:1	1189	0.97	5.55
Total trans C18:1	1176	0.92	3.57
C18:2 *c*-9, 12	1172	0.81	2.30
C18:2 *c*-9, *t*-11	1154	0.85	2.59
Total C18:2	1166	0.75	2.00
C18:3 *c*-9,12,15	1169	0.77	2.11
Total trans C18	1181	0.92	3.59
SFA	1176	1.00	15.34
UFA	1179	0.98	7.62
MUFA	1180	0.98	7.18
PUFA	1180	0.85	2.56
SCFA	1185	0.96	5.10
MCFA	1187	0.98	7.53
LCFA	1188	0.97	5.96
$n-3$	1172	0.77	2.11
$n-6$	1167	0.76	2.03
BCFA	1166	0.85	2.61

[a] The number of samples included in the calibration equation.
[b] Cross validation coefficient of determination.
[c] The ratio of SD to SECV.

cows sampled in the same herd on the same sample date; b_4,b_5, b_6, b_7, and b_8 were the fixed regression coefficients on, respectively, HF_m, MRY_n, DF_o, G_p, JER_q, which were the expected percentages of genes belonging to each of those breeds; b_9 was the fixed regression on HET_r, which was the estimated percentage of heterosis; b_{10} was the fixed regression on REC_s, which was the estimated percentage of recombination loss effect; a_t was the random additive genetic effect of cow t; pe_t was the random permanent environmental effect of cow t, and $e_{ijklmnopqrstu}$ is the random residual effect. HET was calculated as function of the degree of heterozygosity of animals and REC was derived from the heterozygosity of parental gametes which calculations are both described by Van Der Werf and De Boer (1989).

Heritabilities for all traits were calculated separately for MRY and HF using the obtained estimated variance components. The heritability was calculated as:

$$h^2 = \frac{\sigma_a^2}{\sigma_a^2 + \sigma_{pe}^2 + \sigma_e^2},$$

To evaluate the difference between the additive genetic variance components of HF and MRY a log likelihood ratio test was performed using following formula:

$$D = -2\ \ln(\ell R2/\ell R1),$$

where $\ell R1$ is the likelihood for the model to estimate the genetic variance for MRY and $\ell R2$ is the likelihood for the same model for MRY, except in this case the additive genetic variance was fixed at the value of the additive genetic variance of HF. The additive genetic variance for MRY was considered to be significantly different from the value for HF when the test statistic was above the 5% critical value of 2.71 from a mixture of the χ^2 distribution with 0 and 1 degrees of freedom (Self and Liang, 1987). Significance was assessed from the χ^2 distribution with 1 degrees of freedom, which was used for convenience, instead of *P*-values from the required mixture of χ^2 distribution with 0 and 1 degrees of freedom. The results, however, gives a correct representation of the additive genetic variances which are significantly different from each other ($P \leq 0.05$) and which are not ($P > 0.05$).

The additive genetic variances and heritabilities were estimated using a dataset from commercial herds including purebred and crossbred cows. The crossbred cows included animals that are registered having at least 50% genes of MRY or of HF origin. Crossbred animals were included to have as many farms using the MRY breed in the analyses as possible.

3. Results

3.1. Production traits

Additive genetic variances and heritabilities of 5 production traits were estimated for a Dutch MRY population and a Dutch HF population (Table 3). The estimated additive genetic variances of the milk fat percentage and the milk protein percentage were significantly higher (both $P < 0.001$) for the Dutch HF population compared to the Dutch MRY population. In addition, the heritabilities of the milk fat percentage and the milk protein percentage were also higher for the Dutch HF population. The relative differences of the additive genetic variances of the milk fat percentage between both populations was highest, of which the additive genetic variance of HF was with an estimated value of 0.210, 66% higher than those of MRY with 0.072.

3.2. Individual FA C4:0 to C18:0

For all individual FA C4:0 to C18:0 including the unsaturated FA C14:1 *c*-9 and C16:1 *c*-9 the additive genetic variances and heritabilities were estimated for MRY and HF (Table 3). For both, the additive genetic variances as well as the heritabilities, the estimates were lower for MRY compared to HF. Except for the FA 18:0 ($P = 0.1681$), the differences of the estimated additive genetic variances between MRY and HF were significant (C4:0 $P < 0.01$; C6:0-C16:0 $P < 0.001$), where the differences ranged from 42% to 65% relative to the additive genetic variances estimated for HF.

Table 3
The heritability and additive genetic variance of 5 production traits, 14 individual fatty acids and 14 groups of fatty acids for the breeds MRY and HF and the *t*-values of the differences between the MRY and HF additive genetic variances.

	MRY[a]		HF[a]		% of diff. [d,e]
Traits	h^{2}[b]	Var A	h^{2}[c]	Var A	
Production traits					
Milk yield (kg)	0.22	4.2284	0.21	6.4129	34
Fat%	0.22	0.0719	0.49	0.2104	66***
Protein%	0.27	0.0208	0.48	0.0429	52***
Fat yield (kg)	0.17	0.0078	0.16	0.0106	27
Protein yield (kg)	0.16	0.0038	0.17	0.0054	30
Traits g/dL milk					
C4:0	0.28	7.4E−05	0.39	0.000126	42**
C6:0	0.27	4.4E−05	0.50	0.000100	56***
C8:0	0.28	2.3E−05	0.49	0.000051	54***
C10:0	0.29	0.00018	0.48	0.000352	50***
C12:0	0.28	0.00027	0.47	0.000571	52***
C14:0	0.27	0.00162	0.52	0.003813	58***
C14:1 *c*-9	0.27	1.9E−05	0.48	0.000045	59***
C16:0	0.34	0.01842	0.51	0.038513	52***
C16:1 *c*-9	0.17	3.6E−05	0.41	0.000104	65***
C18:0	0.23	4E−06	0.28	0.000010	29***
C18:1 *c*-9	0.1	0.00167	0.17	0.002339	42
Total cis C18:1	0.1	0.00246	0.17	0.004235	40
Total trans C18:1	0.2	0.0028	0.16	0.004646	−18
C18:2 *c*-9, 12	0.26	0.00019	0.26	0.000159	18
C18:2 *c*-9, *t*-11	0.21	0.00346	0.11	0.005174	−78*
Total C18:2	0.22	0.00003	0.34	0.000037	39*
C18:3 *c*-9,12,15	0.17	0.00002	0.31	0.000011	47*
Total trans C18	0.21	4.1E−05	0.16	0.000067	−27
SFA	0.28	2E−06	0.53	0.000004	61***
UFA	0.11	0.00029	0.22	0.000229	51*
MUFA	0.11	0.05544	0.22	0.141201	51*
PUFA	0.21	0.00496	0.19	0.010186	1
SCFA	0.27	0.00407	0.50	0.008307	57***
MCFA	0.28	0.00017	0.53	0.000172	62***
LCFA	0.14	0.00099	0.21	0.002300	37
n−3	0.21	0.0363	0.33	0.094703	44*
n−6	0.22	0.011	0.35	0.017551	34*
BCFA	0.25	5E−06	0.36	0.000009	36*

[a] MRY = Meuse-Rhine-Yssel and HF = Holstein Friesian.
[b] With a SE of on average 0.05 (which was between 0.03 and 0.06) for all estimated heritabilities.
[c] With a SE of 0.01 for all estimated heritabilities.
[d] The difference of the additive genetic variance of MRY compared to the additive genetic variance of HF as percentage of the additive variance of HF.
[e] The *P*-values obtained from the χ^2 distribution with 1 degrees of freedom: *$P < 0.05$; **$P < 0.01$; ***$P < 0.001$.

3.3. C18 UFA

The estimated additive genetic variances and heritabilities of the individual C18 unsaturated FA and groups of these FA are also shown in Table 3. The additive genetic variances and heritabilities of the traits C18:1 *c*-9, Total cis C18:1, Total C18:2, and C18:3 *c*-9, 12, 15 were estimated to be lower for MRY compared to HF. For Total C18:2 and C18:3 *c*-9, 12, 15 the differences of the additive genetic variances between MRY and HF were significant (both $P < 0.05$) with a difference of respectively 39% and 47% relative to the additive genetic variance of HF. The additive genetic variances and heritabilities of the traits Total trans C18:1, C18:2 *c*-9, *t*-11, and Total trans C18 were, however, estimated to be higher for MRY compared to HF. For C18:2 *c*-9, *t*-11 the differences of the additive genetic variances between MRY and HF were significant ($P < 0.05$) with a difference of −78% relative to the additive genetic variance of HF.

3.4. Groups of FA

The estimated additive genetic variances and heritabilities of 10 groups of FA are shown in the bottom part of Table 3 (group definitions are given in Table 1). For both, the additive genetic variances as well as the heritabilities, the estimates were lower for MRY compared to HF, except for the group of FA PUFA. For PUFA, the heritability of MRY was estimated to be 0.21 and for HF 0.19, however, the additive genetic variance was almost similar. For all other groups of FA, except LCFA, the additive genetic variances were significant higher for HF ($P < 0.05$) with a difference ranging from 34% to 62% relative to the additive genetic variance of HF.

4. Discussion

This paper reports estimates of the additive genetic variances and the heritabilities of the detailed FA composition in milk for the Dutch MRY in comparison with the estimates for the HF breed. Analyses were based on predicted FA composition, using MIRS spectra collected from a large number of milk samples. The FA predictions used to predict the additive genetic variances and heritabilities were expressed on milk basis (g of FA/dL of milk) thus the estimated variance components indicate to what extent selection is possible on FA composition as contents of individual FA in milk. Prediction on milk basis was used, because the accuracy of prediction is considerably higher compared to fat basis (g/100 g fat), as shown by De Marchi et al. (2011), Soyeurt et al. (2011), and Rutten et al. (2009). For the present study the RobustMilk calibration equations were used because these equations were developed using data of different breeds including MRY and HF in the Walloon region of Belgium and including multiple countries (Soyeurt et al., 2011) to enlarge the data variability which is essential for the application of MIRS (De Marchi et al., 2014). The predictive ability of these calibration equations in milk of different cattle breeds in the Netherlands, including MRY, has been investigated previously using an independent dataset with both MIRS spectra and gas chromatography measurements (Maurice-Van Eijndhoven et al., 2012). In that study, the predictive ability was evaluated for 10 individual FA and 3 groups of FA,

which are also included in the current study, and the coefficient of determination of the predictions ranged from 0.64 to 1.00. Some descriptive statistics of the calibration equations used in current study are also given in Table 2. Highest predictability, e.g. coefficient of determination close to one and highest RPDs, is especially shown for the predictions of the saturated short and medium FA. To use variability of FA composition within breeds for breeding purposes at the population level, predicted individual FA composition does not necessarily have to be 100% accurate. Indeed, Rutten et al. (2010) showed that the MIRS calibration equations have to be based on a large number of calibration samples, roughly 1,000 samples or more, to optimise the variability of calibration data in order to minimise the loss in potential genetic gain when using predicted FA from MIRS. The calibration equations used in the present study were based on 1,236 samples from multiple breeds and countries to cover a wide range of FA variation. Lowest accuracies for the calibration equations were found for the poly-unsaturated FA (ranging from 0.75 to 0.97) which can lead to bias of the prediction, therefore, results of these FA have to be interpreted carefully. For most of the FA considered in our study, however, it is expected that our results are hardly affected due to the use of MIRS FA predictions, which is confirmed by the fact that the obtained results for HF are generally in line with results published in the literature.

The HF breed is dominating in the Dutch dairy industry which is the main reason explaining the difference in number of records used in this study between MRY and HF. An important question is whether the difference in numbers of records for the MRY versus HF breeds could have contributed to the observed differences in estimated variance components. To examine this, the data of the HF herds were randomly divided in 75 subsets each having approximately the same size as the MRY dataset used in our study. Of these 75 subsets, 6 randomly chosen subsets were used to estimate the heritabilities and additive genetic variances for fat percentage, 4 individual FA, and 1 group of FA (SFA) (Table 4). Estimates for those individual subsets were on average clearly different from the estimates obtained using the MRY dataset, while they were close to the estimates obtained using the large HF dataset. Based on these results of the HF subsets, it was concluded that the differences in results observed between MRY and HF were not due to the differences in the size of the datasets.

It is well known that the FA composition in milk is affected by both genetics as well as the cows diet. Lowest additive genetic variances are found for FA with lower average contents in milk, which are mainly the unsaturated FA. The FA C4:0–C14:0 arise in milk mainly from de novo synthesis (Bauman and Griinari, 2003). This means that a considerable part of the variation in production of these FA is expected to have a genetic origin. In this study, the heritabilities for HF range from 0.39 to 0.52 for the traits C4:0–C14:0 while the heritabilities for MRY range from 0.27 to 0.29. The relative differences between the estimated additive genetic variances of HF and the additive genetic variances of MRY were even larger, and those differences were also highly significant. Soyeurt et al. (2007) reported almost similar heritabilities for C12:0 and C14:0 (respectively 0.29 and 0.31) compared to the estimates for MRY in our study (respectively 0.28 and 0.27). In their study, altogether heritabilities were found ranging from 0.05 to 0.38. Their dataset contained 7 breeds, including animals of the HF (45.39% of the studied population) and MRY (4.31% of the studied population) breeds in Belgium, and also MIRS spectra were used to predict FA composition although they were expressed on fat basis (g/100 g fat). Heritabilities for the Dutch HF population were also estimated by Stoop et al. (2008), which reported heritabilities ranging from 0.22 to 0.71. For the traits C4:0–C14:0 they reported heritabilities ranging from 0.42 to 0.71, which is somewhat higher than in our study (range from 0.39 to 0.52). Compared to C4:0–C14:0, C16:0 is different in the sense that it arises in milk both through de novo synthesis and by uptake of blood circulation (Bauman and Griinari, 2003). The heritabilities for C16:0 were estimated to be (almost) highest compared to the other heritabilities of the same breed in our study, which was in line with Soyeurt et al. (2007). For the individual FA C4:0–C16:1 our results were not in agreement with those of Bobe et al. (2008), however, for these FA their results were generally in disagreement with other results in the literature. The

Table 4
The heritabilities with standard errors and additive genetic variances estimated based on 6 subgroups of HF data.

Traits	HF Group[a,b]						
	1	2	3	4	5	6	Average
Fat%							
h^2	0.51	0.38	0.48	0.42	0.49	0.45	0.46
SE	0.06	0.07	0.07	0.07	0.07	0.07	0.07
Var A	0.209042	0.154326	0.195856	0.178338	0.200369	0.208496	0.191071
C6:0							
h^2	0.48	0.36	0.47	0.46	0.55	0.46	0.46
SE	0.06	0.07	0.07	0.07	0.07	0.07	0.07
Var A	0.000094	0.000070	0.000092	0.000092	0.000110	0.000099	0.000093
C16:0							
h^2	0.49	0.47	0.46	0.41	0.47	0.46	0.46
SE	0.07	0.07	0.07	0.07	0.07	0.07	0.07
Var A	0.033271	0.033076	0.033156	0.030624	0.034186	0.036033	0.033391
C16:1 *c*-9							
h^2	0.53	0.31	0.38	0.34	0.43	0.42	0.40
SE	0.06	0.06	0.07	0.06	0.07	0.07	0.07
Var A	0.000138	0.000074	0.000090	0.000087	0.000108	0.000108	0.000101
C18:1 *c*-9							
h^2	0.28	0.07	0.13	0.13	0.12	0.08	0.14
SE	0.06	0.04	0.06	0.05	0.05	0.05	0.05
Var A	0.007265	0.001499	0.003050	0.003315	0.002962	0.002217	0.003385
SFA							
h^2	0.50	0.46	0.51	0.47	0.53	0.50	0.50
SE	0.07	0.07	0.07	0.07	0.07	0.07	0.07
Var A	0.122625	0.114805	0.129838	0.123965	0.132874	0.141525	0.127605

[a] HF=Holstein Friesian.
[b] Each group is a random representation of the total dataset with record of HF cows and the number of records within each group is ranging from 1823 to 2214.

long chain (more than 16 carbons) unsaturated FA in milk are mainly obtained by uptake from blood circulation, however, also the rumen biohydrogenation and the Δ^9-desaturase activity contribute to the milk content of these FA (Neville and Picciano, 1997). Estimated heritabilities for these FA were lower for both the HF and the MRY breed (heritabilities ranging respectively from 0.11 to 0.34 for HF and 0.10 to 0.26 for MRY) than previously discussed FA. The heritabilities and additive genetic variances were higher for HF compared to MRY except for Total trans C18:1, C18:2 *c*-9, *t*-11, and Total trans C18. However, only the differences in additive genetic variances for C18:2 *c*-9, *t*-11 was significant. Karijord et al. (1982) and Stoop et al. (2008) also reported somewhat lower heritabilities for the long chain and unsaturated FA, which implies a relatively larger influence of environmental aspects compared to genetics on the differences between individuals. In conclusion, although in the other studies FA composition was analysed on fat basis, the relative differences in heritability across FA within other studies tended to be the same as those found in our study.

A possible explanation of (a part of) the differences in additive genetic variances and heritabilities between the MRY and HF breed suggests that the genetic architecture differs between those breeds, i.e. differences in genomic variation. The lower genetic variance for MRY was nevertheless unexpected, as there has been relatively very little emphasis on selection for high-input high-output production systems in MRY compared to HF over the last decades. The FA composition of bovine milk is known to be effected by several genes with a moderate to large effect. Two genes, having polymorphisms with reported effects on FA composition, are *DGAT1* and SCD1 (e.g. Thaller et al., 2003; Mele et al., 2007; Schennink et al., 2008). Polymorphisms in those genes affect the production of FA and, thus, also the genetic variances within breeds as there are different genotypes. Especially the *DGAT1* K232A polymorphism is reported having significant effects on the milk production traits and some medium chain SFA and long chain UFA (Schennink et al., 2007). Percentages of the genetic variances of FA composition in milk explained by the *DGAT1* K232A polymorphism were reported ranging from 1% up to 53% within the HF breed in the Netherlands (Schennink et al., 2007, 2008). Those high percentages of explained genetic variance, are due to the high minor allele frequency of *DGAT1*, i.e. Schennink et al. (2007) reported a frequency of 0.40 for the 232 K allele in the Dutch HF population. Considering what is known about the *DGAT1* K232A polymorphism, the lower genetic variances in MRY for a number of FA may be because one of the alleles at the *DGAT1* locus has an extreme frequency such that the contribution of *DGAT1* to the genetic variance in MRY is limited. Because the allele frequency of the *DGAT1* K232A polymorphism is currently not known for the MRY breed, this hypothesis will be tested in future research.

5. Conclusion

For both, MRY and HF, additive genetic variances and heritabilities were estimated for detailed FA composition in milk. The additive genetic variances as well as the heritabilities for the SCFA and MCFA, which mainly arise in milk by de novo synthesis, were generally lower for MRY than for HF. Lower heritabilities and less significant differences in heritability between the breeds were estimated for the long chain C18 FA that are mainly obtained by uptake from blood circulation. Lower variances in MRY may be because of a difference of their genetic architecture compared to HF. In conclusion, our results show that the HF breed has substantially larger genetic variance for most FA compared to MRY, despite its stronger selection for milk yield traits in the past. As the estimated genetic variances for MRY were clearly lower, and because it is know that the *DGAT1* locus has an intermediate allele frequency in HF, it is hypothesised that the *DGAT1* locus has a more extreme minor allele frequency in the Dutch MRY population, which will be tested in future research.

Acknowledgements

This study was financially supported by the Ministry of Economic Affairs (Programme “Kennisbasis Research”, code: KB-04-002-021 and KB-05-003-041). CRV BV (Arnhem, The Netherlands) is thanked for the contribution of the data including mid-infrared spectra. The RobustMilk project team is thanked for the use of the calibration equations. The RobustMilk project is financially supported by the European Commission under the Seventh Research Framework Programme, Grant Agreement KBBE-211708. This publication represents the views of the authors, not the European Commission, and the Commission is not liable for any use that may be made of the information. Furthermore, Johan van Arendonk and Henk Bovenhuis (Wageningen University, Animal Breeding and Genomics Centre, Wageningen, The Netherlands) as well as Mari Smits (Wageningen UR Livestock Research, Animal Breeding and Genomics Centre, Wageningen, The Netherlands) and Sipke Joost Hiemstra (Centre for Genetic Resources, the Netherlands (CGN) of Wageningen UR) are thanked for comments on the set-up and data analysis of this study.

References

Bastin, C., Gengler, N., Soyeurt, H., 2011. Phenotypic and genetic variability of production traits in milk fatty acid contents across days in milk for Walloon Holstein first-parity cows. J. Dairy Sci. 94, 4152–4163.
Bauman, D.E., Griinari, J.M., 2003. Nutritional regulation of milk fat synthesis. Annu. Rev. Nutr. 23, 203–227.
Baumgard, L.H., Sangster, J.K., Bauman, D.E., 2001. Milk fat synthesis in dairy cows is progressively reduced by increasing supplemental amounts of trans-10, cis-12 conjugated linoleic acid (CLA). J. Nutr. 131, 1764–1769.
Bobe, J.A., Lindberg, G.L., Freeman, A.E., Beitz, D.C., 2008. Short communication: estimates of genetic variation of milk fatty acids in US Holstein cows. J. Dairy Sci. 91, 1209–1213.
De Marchi, M., Toffanin, V., Cassandro., M., Penasa, M., 2014. Invited review: mid-infrared spectroscopy as phenotyping tool for milk traits. J. Dairy Sci. 97, 1171–1186.
De Marchi, M., Penasa, M., Cecchinato, A., Mele, M., Secchiari, P., Bittante, G., 2011. Effectiveness of mid-infrared spectroscopy to predict fatty acid composition of Brown Swiss bovine milk. Animal 5, 1653–1658.
Dijkstra, J., van Zijderveld, S.M., Apajalahti, J.A., Bannink, A., Gerrits, W.J.J., Newbold, J.R., Perdok, H.B., Berends, H., 2011. Relationships between methane production and milk fatty acid profiles in dairy cattle. Anim. Feed Sci. Technol. 166–167, 590–595.
Gilmour, A.R., Gogel, B.J., Cullis, B.R., Thompson, R., 2009. ASReml User Guide Release 3.0. VSN International Ltd., Hemel Hempstead, UK.
Karijord, O., Standal, N., Syrstad, O., 1982. Sources of variation in composition of milk fat. Z. Tierz. Zuchtungsbiol. 99, 81–93.
Maurice-Van Eijndhoven, M.H.T., Soyeurt, H., Dehareng, F., Calus, M.P.L., 2012. Validation of fatty acid predictions in milk using mid-infrared spectrometry across cattle breeds. Animal 7, 348–354.
Maurice-Van Eijndhoven, M.H.T., Bovenhuis, H., Soyeurt, H., Calus, M.P.L., 2013. Differences in milk fat composition predicted by mid-infrared spectrometry among dairy cattle breeds in the Netherlands. J. Dairy Sci. 96, 2570–2582.
Mele, M., Conte, G., Castiglioni, B., Chessa, S., Macciotta, N.P.P., Serra, A., Buccioni, A., Pagnacco, G., Secchiari, P., 2007. Stearoyl-coenzyme A desaturase gene polymorphism and milk fatty acid composition in Italian Holsteins. J. Dairy Sci. 90, 4458–4465.
Mensink, R.P., Zock, P.L., Kester, A.D.M., Katan, M.B., 2003. Effects of dietary fatty acids and carbohydrates on the ratio 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.
Neville, M.C., Picciano, M.F., 1997. Regulation of milk lipid secretion and composition. Annu. Rev. Nutr. 17, 159–183.
Palmquist, D.L., Stelwagen, K., Robinson, P.H., 2006. Modifying milk composition to increase use of dairy products in healthy diets-Preface. Anim. Feed Sci. Technol. 131, 149–153.
Rutten, M.J.M., Bovenhuis, H., van Arendonk, J.A.M., 2010. The effect of the number of observations used for Fourier transform infrared model calibration for bovine milk fat composition on the estimated genetic parameters of the predicted data. J. Dairy Sci. 93, 4872–4882.

Rutten, M.J.M., Bovenhuis, H., Hettinga, K.A., van Valenberg, H.J.F., van Arendonk, J. A.M., 2009. Predicting bovine milk fat composition using infrared spectroscopy based on milk samples collected in winter and summer. J. Dairy Sci. 92, 6202–6209.
Schennink, A., Heck, J.M.L., Bovenhuis, H., Visker, M.H.P.W., van Valenberg, H.J.F., van Arendonk, J.A.M., 2008. Milk fatty acid unsaturation: Genetic parameters and effects of stearoyl-CoA desaturase (SCD1) and acyl CoA: diacylglycerol acyltransferase 1 (DGAT1). J. Dairy Sci. 91, 2135–2143.
Schennink, A., Stoop, W.M., Visker, M.H.P.W., Heck, J.M.L., Bovenhuis, H., van der Poel, J.J., van Valenberg, H.J.F., van Arendonk, J.A.M., 2007. DGAT1 underlies large genetic variation in milk-fat composition of dairy cows. Anim. Genet. 38, 467–473.
Self, S.C., Liang, K.Y., 1987. Asymptotic properties of maximum likelihood estimators and likelihood ratio tests under non-standard conditions. J. Am. Stat. Assoc. 82, 605–610.
Smet, K., De Block, J., De Campeneere, S., De Brabander, D., Herman, L., Raes, K., Dewettinck, K., Coudijzer, K., 2009. Oxidative stability of UHT milk as influenced by fatty acid composition and packaging. Int. Dairy J. 19, 372–379.
Stull, J.W., Brown, W.H., Valdez, C., 1966. Fatty acid composition of milk. Part III. Variation with stage of lactation. J. Dairy Sci. 49, 1401–1405.
Soyeurt, H., Dehareng, F., Gengler, N., McParland, S., Wall, E., Berry, D.F., 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.
Soyeurt, H., Gillon, A., Vanderick, S., Mayeres, P., Bertozzi, C., Gengler, N., 2007. Estimation of heritability and genetic correlations for the major fatty acids in bovine milk. J. Dairy Sci. 90, 4435–4442.
Sterk, A., Johansson, B.E.O., Taweel, H.Z.H., Murphy, M., van Vuuren, A.M., Hendriks, W.H., Dijkstra, J., 2011. Effects of forage type, forage to concentrate ratio, and crushed linseed supplementation on milk fatty acid profile in lactating dairy cows. J. Dairy Sci. 94, 6078–6091.
Stoop, W.M., van Arendonk, J.A.M., Heck, J.M.L., van Valenberg, H.J.F., Bovenhuis, H., 2008. Genetic parameters for major milk fatty acids and milk production traits of Dutch Holstein-Friesians. J. Dairy Sci. 91, 385–394.
Thaller, G., Kramer, W., Winter, A., Kaupe, B., Erhardt, G., Fries, R., 2003. Effects of DGAT1 variants on milk production traits in German cattle breeds. J. Anim. Sci. 81, 1911–1918.
Van Der Werf, J.H.J., De Boer, W., 1989. Estimation of genetic parameters in a crossbred population of black and white dairy cattle. J. Dairy Sci. 72, 2615–2623.
Wilmink, J.B.M., 1987. Adjustment for test-day milk, fat, and protein yield for age, season and days-in-milk. Livest. Prod. Sci. 16, 335–348.