

# Relationship between proxies of energy states and nitrogen use efficiency for Holstein cows in early lactation

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## Abstract

The purposes of this study were to estimate the genetic parameters of the energy status indicator (C18:1 *cis*-9) of Holstein cows in early lactation and its relationship with nitrogen use efficiency (NUE) (predicted NUE and milk urea concentration). After editing, the data included 143,517 records within 5 to 50 days in milk from 52,198 cows, and 3,546 animals in the pedigree with 28,427 SNPs. Two multiple-trait repeatability models were used in this study. In early lactation, the average C18:1 *cis*-9 was gradually decreasing and was highest in May. The heritabilities of C18:1 *cis*-9 for primiparous and multiparous cows were 0.12 and 0.09, respectively. The C18:1 *cis*-9 had positive genetic correlations with predicted NUE (from 0.28 to 0.67), and weak genetic correlations with milk yield, milk urea concentration (from -0.15 to 0.14). This study suggests that breeding for NUE alone may enhance energy troubles in early lactation.

## Introduction

Most dairy cows will be in a negative energy balance (NEB) in early lactation due to energy intake that cannot meet the needs of milk production (Churakov et al., 2021). If the dairy cow is in NEB state for too long, the risk of the dairy cow suffering from metabolic diseases (such as ketosis) will increase (Zachut et al., 2020), but it is difficult to directly detect the energy status of dairy cows. Usually, people use indirect prediction of the NEB of dairy cows as an alternative method. Although using milk mid-infrared (MIR) spectra to indirectly predict the NEB of dairy cows is a good method, the coefficient of determination of its prediction model is varying in different studies (from 0.48 to 0.77) (Ho et al., 2019; Smith et al., 2019).

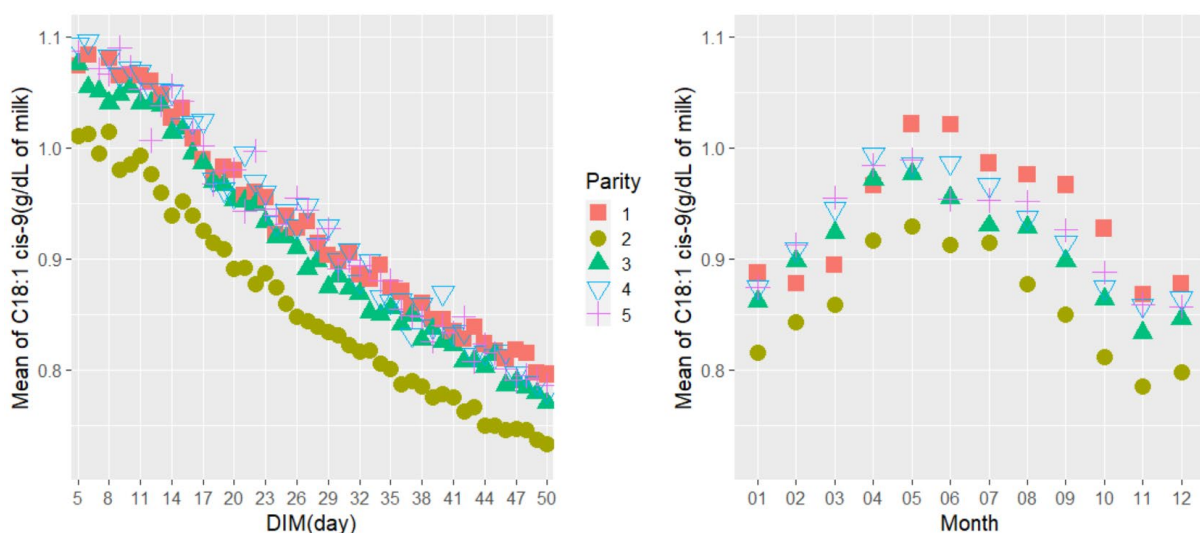
Studies have shown that oleic acid (C18:1 *cis*-9) is a good proxy for identifying NEB in dairy cows (Churakov et al., 2021, Guinguina et al., 2021). When the cow is in NEB, its body fat reserves will be mobilized, leading to an increase in C18:1 *cis*-9 in milk (Zachut et al., 2020). The C18:1 *cis*-9 can be accurately and quickly predicted using MIR. The determination coefficients for cross-validation of its prediction model were 0.95 (Soyeurt et al., 2011), which was confirmed by the study of Grelet et al. (2014). The genetic parameters of C18:1 *cis*-9 have been reported but they only focus on the first parity (Bastin et al., 2011; Mehtiö et al., 2020). As the nitrogen use efficiency (NUE) of dairy cows has attracted attention (Chen et al., 2021b) in recent years, we guess that its addition to breeding may increase the NEB of dairy cows. In this study, the new proxy (predicted NUE, PNUE) and traditional proxy (milk urea concentration, MU) of NUE were used as representatives to evaluate their relationships with C18:1 *cis*-9 (Chen et al., 2021a, 2021b). At the same time, this research has added milk yield (MY) as a representative of the main economic traits.

The purposes of this study were to estimate the genetic parameters of C18:1 *cis*-9 in the early lactation of the first five parities of dairy cows and its relationship with PNUE, MU, and MY.

## Materials & methods

**Data.** All data were collected from 2012 to 2019 during the official milk recording in the Walloon Region of Belgium. The MIR analysis of milk samples was performed by MilkoScan

FT 6000 and FT+ spectrometers (Foss Electric, Hillerød, Denmark) to provide MIR spectra and milk composition. Edits excluded irregular data for daily MY (<1 and >99 kg) and MU (<2 and >70 mg/dL). The filtering method for C18:1 *cis*-9 (g/dL of milk) and PNUE was the same as Chen et al. (2021b). Furthermore, the range of days in milk (DIM) was from 5 to 50, parity within 5, and the ranges of accepted calving age were: from 23 to 37, 35 to 52, 47 to 67, 59 to 83, 71 to 96 months for the first to fifth parity, respectively. After filtering, 134,517 records from 52,198 Holstein cows on 774 farms were used in this study. The pedigree related to the dataset comprised 120,933 animals, and 3,546 (397 bulls) of them had genotypes with 28,427 SNPs. The genotype data were provided by the cattle breeding system in the Walloon Region of Belgium. Besides, each trait was divided into primiparous and multiparous (including second to fifth parity) classes, except the MU (based on the research results of Chen et al., 2021a). Hereafter, C18:1 *cis*-9, PNUE, and MY data were divided following parities and identified as C18:1 *cis*-9\_1, PNUE1, MY1, C18:1 *cis*-9\_2+, PNUE2+, and MY2+. At the same time, based on its trend with DIM (Figure 1), C18:1 *cis*-9 was also divided into 3 traits, C18:1 *cis*-9\_1 (first parity), C18:1 *cis*-9\_2 (second parity), C18:1 *cis*-9\_3+ (from third to fifth parity). All milk MIR spectra were standardized according to Grelet et al. (2015). The C18:1 *cis*-9 and PNUE of each cow were predicted by the equations based on the models established by Grelet et al. (2014, 2020). The determination coefficients and root mean square errors of cross-validation were 0.95 and 0.06% for C18:1 *cis*-9, and 0.68 and 5.01% for the PNUE model, respectively.



**Figure 1. The curves of the mean of C18:1 *cis*-9 (g/dL of milk) with days in milk (DIM) and month for Holstein cows in the first five parities.**

**Models.** The three-trait and seven-trait repeatability models were used to estimate the variance components for C18:1 *cis*-9 (C18:1 *cis*-9\_1, C18:1 *cis*-9\_2, and C18:1 *cis*-9\_3+) and seven traits (C18:1 *cis*-9\_1, PNUE1, MY1, MU, C18:1 *cis*-9\_2+, PNUE2+, and MY2+). The models were the same as Chen et al. (2021b), expect the number of traits.

**Genetic parameters.** The variance components, heritability ( $h^2$ ), and genetic correlations of the three-trait model were estimated by AIREMLF90 (version 1.149). The variance components of the seven-trait model were estimated by Gibbs sampling in THRGIBBS1F90 (version 2.119), and posterior convergence was analyzed by POSTGIBBSF90 (version 3.15). Among them, the posterior means of variances were obtained using 100,000 samples after a burn-in of 20,000 samples. Using the mean of the posterior variance components, the  $h^2$ , genetic, and phenotypic

correlations were calculated. Approximate standard errors (SE) of parameters were obtained from POSTGIBBSF90 (version 3.15) program.

## Results & discussion

**Descriptive phenotype.** The average (standard deviation, SD) C18:1 *cis*-9 in primiparous and multiparous cows was 0.92 g/dL (0.25) and 0.89 g/dL (0.24), respectively. This was in line with other studies (Bastin et al., 2011; Mehtiö et al., 2020; Churakov et al., 2021). The trends of the average C18:1 *cis*-9 of dairy cows in early lactation were shown in Figure 1. The average C18:1 *cis*-9 gradually decreased from DIM 5 to 50, ranging from 0.73 (SD: 0.15) to 1.11 (SD: 0.28), which is consistent with the report by Churakov et al. (2021). Within the months of the year, the average C18:1 *cis*-9 ranged from 0.78 (SD: 0.20) to 1.02 (SD: 0.25), and each parity reached its maximum in May, except for the fourth parity. It may be that C18:1 *cis*-9 was very sensitive to heat stress in dairy cows (Hammami et al., 2014), and cows got fresh pasture in May (Frétin et al., 2019).

**Heritabilities.** In the seven-trait model, the  $h^2$  (SE) of C18:1 *cis*-9 in primiparous and multiparous were 0.12 (0.01) and 0.09 (0.01) (Table 1), which were consistent with our results in the three-trait model (not shown). The  $h^2$  of C18:1 *cis*-9 in this study was similar to the results obtained in the same area of the previous study (Bastin et al., 2011), and was also consistent with the results obtained in the Nordic Red Dairy cattle study in Finland (Mehtiö et al., 2020). Moreover, the  $h^2$  of PNUE and MU were consistent with the results of our previous studies (Chen et al., 2021a, 2021b).

**Table 1. Heritabilities<sup>1</sup> (diagonal, bold), genetic correlations (above the diagonal) and phenotypic correlations (below the diagonal) among the four features<sup>2</sup> for primiparous (n=41,035) and multiparous (n=93,482) Holstein cows in early lactation.**

	C18:1 <i>cis</i> -9 1	PNUE1	MY1	MU	C18:1 <i>cis</i> -9 2+	PNUE2+	MY2+
C18:1 <i>cis</i> -9_1	<b>0.12</b>	0.67	0.14	0.03	0.67	0.36	0.00
PNUE1	0.33	<b>0.17</b>	0.71	0.12	0.28	0.70	0.50
MY1	0.10	0.47	<b>0.18</b>	0.01	-0.14	0.51	0.78
MU	0.00	0.03	0.01	<b>0.12</b>	-0.15	0.04	-0.02
C18:1 <i>cis</i> -9_2+	0.09	0.03	-0.04	0.02	<b>0.09</b>	0.48	-0.05
PNUE2+	0.07	0.15	0.13	0.03	0.34	<b>0.12</b>	0.74
MY2+	0.02	0.15	0.21	0.03	0.06	0.58	<b>0.13</b>

<sup>1</sup>: the standard errors of all heritabilities, genetic correlations, and phenotypic correlations are 0.01, less than 0.07, and less than 0.01, respectively.

<sup>2</sup>: the four features were divided into seven traits, namely C18:1 *cis*-9 (g/dL of milk) (C18:1 *cis*-9\_1), predict nitrogen use efficiency (%) (PNUE1), daily milk yield (kg) (MY1) in primiparous cattle, and milk urea concentration (mg/dL) (MU) in the first five parities, and C18:1 *cis*-9\_2+, PNUE2+, MY2+, corresponding to multiparous cattle.

**Correlations.** The genetic and phenotypic correlations of the seven traits were described in Table 1. From the three-trait model analysis, the genetic correlations of the first and second with third parity+ traits were 0.70 and 0.93, respectively. Then we changed it to two traits, namely primiparous as the first and multiparous as the second one. The genetic correlations between C18:1 *cis*-9 and PNUE were positive (from 0.28 to 0.67), which indicates that increasing NUE in the early lactation may be detrimental to its energy balance. Another reason is that in early lactation, cows both mobilize energy and protein body reserves, so NUE is artificially high. This was similar to the results of Hurley et al. (2018) who showed that improving feed efficiency was not conducive to the energy balance of cattle. The C18:1 *cis*-9 had weak genetic correlations with MY and MU, ranging from -0.14 to 0.14, and from -0.15 to

0.03, respectively. The phenotypic correlations between C18:1 *cis*-9 and PNUE were moderately positive in the same parity class (0.33 and 0.34). However, the C18:1 *cis*-9 had low phenotypic correlations with MY and MU, ranging from -0.04 to 0.10.

### Conclusions

The C18:1 *cis*-9 had low  $h^2$  in the early lactation of the first five parities for Holstein cows. In addition, the C18:1 *cis*-9 had positive genetic correlations with PNUE (unfavorable), and low genetic correlations with MY and MU (favorable). This study suggests that breeding for NUE alone may enhance energy troubles in early lactation.

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