

## Genetic parameters of butter softness and spreadability as new traits in Dual-Purpose Belgian Blue

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### Abstract

The Belgian Blue (BB) breed, originated from central and upper Belgium in the 19<sup>th</sup> century, is composed of two strains including Beef Belgian Blue (BBB) and Dual-Purpose Belgian Blue (DPBB). The BBB is the most important breed for beef production in Belgium, while the DPBB is the most important local dual-purpose cattle breed reared in dairy farms located in the Walloon Region of Belgium. Through their local identity and resilient characteristics, DPBB cows could be more popular on organic and direct sale-oriented farms in Belgium. In recent decades, human consumption patterns of dairy products have changed, and consumers are more aware of the effects of milk fatty acids (FA) on health. Furthermore, the milk FA composition plays an important role in the quality of butterfat. Therefore, interest in changing the milk FA profile is growing, which motivated researchers to define new traits related to milk FA profile and evaluate the feasibility of including them in selection programs of dairy cattle. In this study, two new traits related to milk FA profile were defined: butter softness (BSO) defined as the ratio of unsaturated to saturated FAs, and butter spreadability (BSP) defined as the ratio of C18:1 (cis-9) FA to C16:0 FA. Data of 69 369 test-day records of milk yield (MY), fat percentage (FP), protein percentage (PP), BSO, and BSP collected from 2007 to 2020 on 7 392 first parity, and 5 185 second parity cows distributed in 104 herds throughout Wallonia were used. Mean heritability estimates across lactation were 0.33 and 0.36 for BSO and 0.24 and 0.32 for BSP in the first and second parity, respectively. Mean genetic correlation estimates between BSO and MY, FP, and PP were, respectively, -0.30, -0.38, and -0.01 in the first and -0.31, -0.46 and -0.12 in the second parity. Mean genetic correlation estimates between BSP and MY, FP, and PP were, respectively, -0.27, -0.38, and -0.06 in the first and -0.27, -0.45 and -0.17 in the second parity. Observed moderate (negative) genetic correlations estimated between the examined traits and milk traits indicate that genetic selection for milk traits would decrease quality of butterfat. High positive genetic correlation was found between BSO and BSP; however, BSO showed a higher heritability than BSP. In addition, the prediction of BSO is more efficient than BSP. Therefore, when a choice must be made between these traits, the inclusion of BSO in selection programs appears to be more justified than BSP.

**Key words:** butter quality, genetic parameter, Dual-Purpose Belgian Blue

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## Introduction

The Belgian Blue (BB) breed originated from central and upper Belgium in the 19<sup>th</sup> century, from crossing local Belgian dairy cattle with Shorthorns (Coopman, 2008). The BB is now composed of two strains including Beef Belgian Blue (BBB) and Dual-Purpose Belgian Blue (DPBB) (Gengler et al., 2007, Mota et al., 2017). The BBB is the most important breed for beef production in Belgium, while the DPBB is the most important local dual-purpose cattle breed reared in dairy farms located in the Walloon Region of Belgium. Smaller populations of DPBB exist in Flanders and also in northern France where they are called “Belgisch-Witblauw Dubbeldoel” and “Bleue du Nord”, respectively. It has been documented that milk produced by DPBB cows has a higher quality than milk produced by high-output international breeds such as Holstein, Jersey, or Brown Swiss (Soyeurt et al., 2006, Bastin et al., 2020). Therefore, DPBB cows could be more popular on organic and direct sale-oriented farms in Belgium.

The fat content of milk is of economic importance because it is a factor affecting milk price in most countries of the world. However, the quality and processing ability of milk are highly correlated with contents and compositions of milk fat. In addition, in recent decades, human consumption patterns of dairy products have changed, and consumers pay more attention to the health aspects of the food. Biologically active lipid substances are primarily saturated fatty acids (SFA), monounsaturated fatty acids (MUFA), and polyunsaturated fatty acids (PUFA). Besides the positive effect of milk unsaturated FAs (UFA) on human health, increasing the proportion of UFA in bovine milk fat has a positive impact on the technological properties of butter (Soyeurt et al., 2008). Therefore, interest in changing the milk fatty acid profile is growing. Moreover, unfavourable changes may happen through correlated responses. All

these facts have motivated researchers to define new traits related to milk fatty acids and to evaluate the feasibility of including them in selection programs of dairy cattle.

The objective of this study was to estimate the genetic parameters for butter softness and butter spreadability as new traits related to milk fatty acid profile in DPBB. Results from this study are potentially useful contributions to the establishment of genetic evaluations for milk quality traits in DPBB.

## Materials and Methods

### *Phenotypic data*

The data used was a subset of the available data and consisted in test-day records of milk yield (MY), fat percentage (FP), protein percentage (PP), and selected FAs contents on Dual-Purpose Belgian Blue (DPBB) cows. Cows with missing birth date, calving date, and parity number were excluded. Only records from the first and second parities that had data for all traits on a given test day were kept. Records from DIM lower than 5 d and greater than 365 d were eliminated. Within cow, if parity two was present, parity one had to be also present. Herds were required to have a minimum of 10 cows in the data set to be included in the analysis. Age at the first calving (AFC) was calculated as the difference between birth date and first calving date and restricted to the range of 640 to 1500 d. Daily MY, FP, and PP were restricted to the range of 1 to 70 kg, 1 to 9 % and 1 to 7 %, respectively. The edited data consisted of 69 369 test-day records of milk yield and milk compositions collected during 2007 to 2020 on 12 577 lactations of 7 392 DPBB cows distributed in 104 herds in the Wallonia region of Belgium. The number of test-day records on the first and second parity cows were, 40 903 (on 7 392 cows) and 28 466 (on 5 185 cows), respectively. Full pedigree consisted of 36 955 females and 6 742 males. Two new traits related to milk fatty acids were defined. Butter softness (BSO) defined as the ratio of

unsaturated to saturated FAs, and butter spreadability (BSP) defined as the ratio of C18:1 (cis-9) FA to C16:0 FA.

**Predicted concentrations of fatty acid in milk.**

All milk samples were analyzed by MIR spectrometer (Foss MilkoScan FT6000; Foss, Hillerød, Denmark), an instrument that also provided the standard milk recording analyses. The methodology used to predict the FA contents was described previously by Soyeurt et al. (2011). Prediction equations used were improved versions (i.e., extended reference data) of those described by Soyeurt et al. (2011) using mid infrared (MIR) milk spectral data that is available for many recent test-days recorded in the Walloon Region of Belgium.

**Genotypic data**

In this study and derived future genetic evaluations, genomic was from the beginning considered. Genotypes were available on 1 699 animals (639 males and 1 060 females). SNP with minor allele frequency (MAF) less than 5%, and those deviated from Hardy-Weinberg equilibrium were excluded. In total, 28,266 SNPs located on the 29 BTAs were used in the genomic analysis.

**Model**

The following random regression (RR) test-day animal model through multiple-trait, multiple-lactation was used to estimate variance components for test-day records of included traits:

$$y_{ijklmn} = \mu + \text{HTD}p_i + \sum_{b=0}^4 AS_j\theta_b(t) + \sum_{b=0}^2 HY_k\theta_b(t) + \sum_{b=0}^2 pe_m\theta_b(t) + \sum_{b=0}^2 a_m\theta_b(t) + e_{ijklmn}$$

where  $y_{ijklmn}$  is the test-day record (MY, FP, PP, and BSO or BSP) on DIM  $n$  of cow  $m$  in parity  $l$ , belonging to  $i^{\text{th}}$  class of HTD $p$ ,  $j^{\text{th}}$  class of AS, and  $k^{\text{th}}$  class of HY, HTD $p_i$  is the fixed effect of  $i^{\text{th}}$  class of herd-test day-parity,

AS $_k$  is the fixed effect of age-season of calving defined as the following: age at calving class (six and four classes of age at calving were created for the first and second parity, respectively)  $\times$  season of calving (four seasons: winter from January to March, spring from April to June, summer from July to September and autumn from October to December),  $\sum_{b=0}^2 HY_k\theta_b(t)$ ,  $\sum_{b=0}^2 pe_m\theta_b(t)$ , and  $\sum_{b=0}^2 a_m\theta_b(t)$  are, respectively, the random regression coefficients of herd-year of calving, permanent environmental, and additive effects,  $e_{ijklmn}$  is the residual effect.

Regression curves were modeled using Legendre polynomials of order 2. The additive genetic, herd-year of calving, permanent environment, and residual variances were obtained as follows:

$$\text{Var} \begin{bmatrix} \mathbf{hy} \\ \mathbf{p} \\ \mathbf{a} \\ \mathbf{e} \end{bmatrix} = \begin{bmatrix} \mathbf{I} \otimes \mathbf{HY} & \mathbf{0} & \mathbf{0} & \mathbf{0} \\ \mathbf{0} & \mathbf{I} \otimes \mathbf{P} & \mathbf{0} & \mathbf{0} \\ \mathbf{0} & \mathbf{0} & \mathbf{Ga} \otimes \mathbf{H} & \mathbf{0} \\ \mathbf{0} & \mathbf{0} & \mathbf{0} & \mathbf{R} \end{bmatrix}$$

Where  $\mathbf{HY}$  is the 24 $\times$ 24 covariance matrix of the herd-year of calving regression coefficients.  $\mathbf{P}$  is the 24 $\times$ 24 covariance matrix of the permanent environmental regression coefficients. Blocks within  $\mathbf{R} = \sum^+ \mathbf{R}^p$  contain diagonal matrices (4 $\times$ 4) of residual covariances among traits with elements that depend on parity ( $\mathbf{p}$ ). Residual covariances among traits on the same testday were therefore allowed to be different from zero, and residual covariances were the same within each parity.  $\mathbf{Ga}$  is the 24 $\times$ 24 covariance matrix of the additive genetic regression coefficients,  $\mathbf{H}$  is a matrix that combines pedigree and genomic relationships, and its inverse consists of the integration of additive and genomic relationship matrices,  $\mathbf{A}$  and  $\mathbf{G}$ , respectively (Aguilar et al., 2010):

$$\mathbf{H}^{-1} = \mathbf{A}^{-1} + \begin{bmatrix} 0 & 0 \\ 0 & \mathbf{G}^{-1} - \mathbf{A}_{22}^{-1} \end{bmatrix}$$

In this equation,  $\mathbf{A}$  is the numerator relationship matrix based on pedigree for all animals;  $\mathbf{A}_{22}$  is the numerator relationship matrix for genotyped animals; and  $\mathbf{G}$  is the weighted genomic relationship matrix obtained using following function:

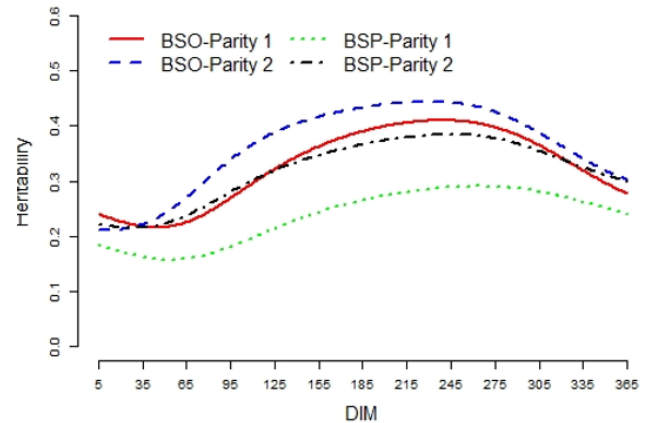
$$\mathbf{G} = \mathbf{G}^* * 0.95 + \mathbf{A}_{22} * 0.05$$

The  $\mathbf{G}^*$  is the genomic relationship matrix obtained using the following function described by VanRaden (2008):

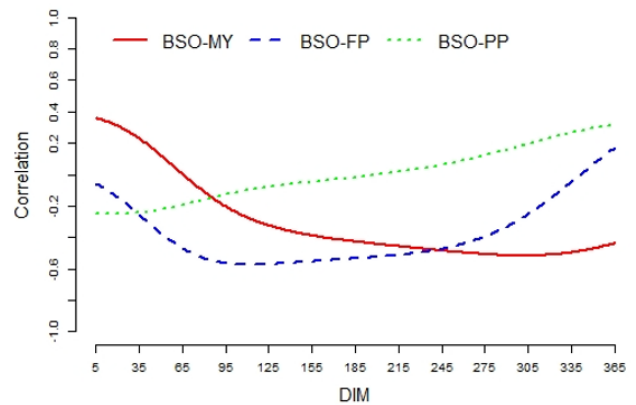
$$\mathbf{G}^* = \frac{\mathbf{ZDZ}'}{\sum_{i=1}^M 2p_i(1 - p_i)}$$

where  $\mathbf{Z}$  is a matrix of gene content adjusted for allele frequencies (0, 1 or 2 for  $aa$ ,  $Aa$ , and  $AA$ , respectively);  $\mathbf{D}$  is a diagonal matrix of weights for SNP variances ( $\mathbf{D} = \mathbf{I}$ );  $M$  is the number of SNP, and  $p_i$  is the minor allele frequency of the  $i^{\text{th}}$  SNP. The  $\mathbf{H}$  matrix was built scaling  $\mathbf{G}$  based on  $\mathbf{A}_{22}$ , considering that the average of diagonal of  $\mathbf{G}$  is equal to the average of diagonal of  $\mathbf{A}_{22}$ , and the average of off-diagonal  $\mathbf{G}$  is equal to average of off-diagonal  $\mathbf{A}_{22}$ .

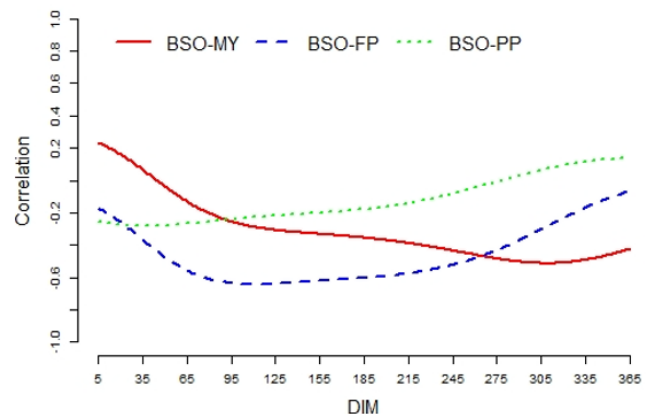
The (co)variance components were estimated by Bayesian inference using the GIBBS3F90 program (Aguilar et al., 2018). Gibbs sampling was used to obtain marginal posterior distributions for the various parameters using a single chain of 100 000 iterates. The first 20 000 iterates of each chain were regarded as a burn-in period to allow sampling from the proper marginal distributions. The length of this burn-in period was determined by visually inspecting plots of sample values across rounds. Genetic (co)variances on each test day were calculated using the equation described by Jamrozik and Schaeffer (1997). Daily heritability was defined as the ratio of genetic variance to the sum of genetic, permanent environmental, herd-year calving, and residual variances at a given DIM.



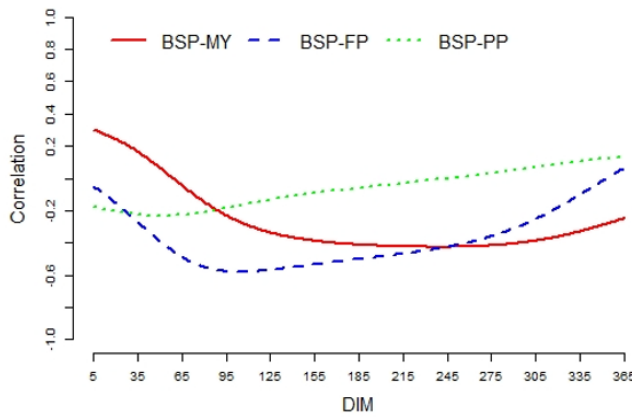
**Figure 1.** Heritability estimates for BSO and BSP in the first (A) and second parity



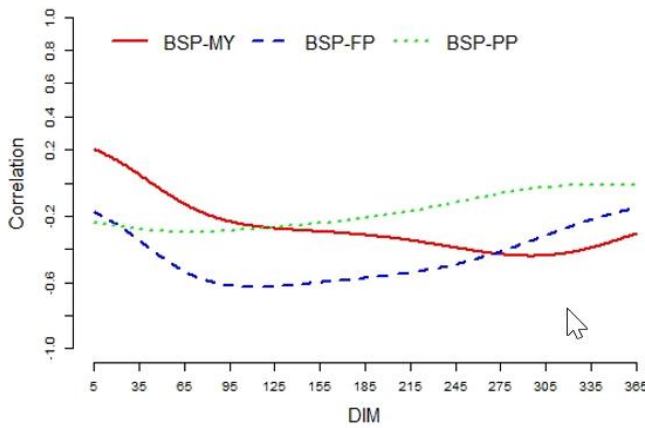
**Figure 2.** Genetic correlation estimates between BSO and milk yield (BSO-MY), fat percentage (BSO-FP), and protein percentage (BSO-PP) in the first parity.



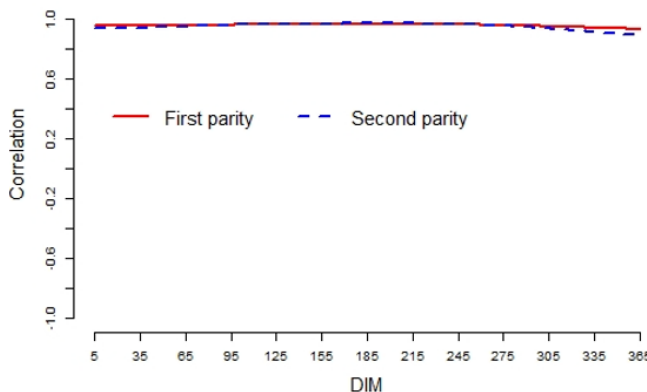
**Figure 3.** Genetic correlation estimates between BSO and milk yield (BSO-MY), fat percentage (BSO-FP), and protein percentage (BSO-PP) in the second parity.



**Figure 4.** Genetic correlation estimates between BSP and milk yield (BSP-MY), fat percentage (BSP-FP), and protein percentage (BSP-PP) in the first parity.



**Figure 5.** Genetic correlation estimates between BSP and milk yield (BSP-MY), fat percentage (BSP-FP), and protein percentage (BSP-PP) in the second parity.



**Figure 6.** Genetic correlation estimates between BSP and BSO in the first and second parity.

## Results & Discussion

Mean (SD) daily milk yield was 12.56 (4.08) kg with 3.71 (0.59) % of fat and 3.34 (0.37) % of protein in the first parity, while the mean (SD) milk yield was 14.69 (5.49) kg with 3.69 (0.63) % of fat and 3.34 (0.38) % of protein in the second parity. Mean (SD) BSO and BSP were, respectively, 0.61 (0.16) and 0.56 (0.16) in the first and 0.59 (0.16) and 0.54 (0.15) in the second parity. The heritability estimated in daily basis for BSO and BSP in the first and second parities are presented in Figure 1. Mean heritability estimates across lactation were 0.33 and 0.36 for BSO and 0.24 and 0.32 for BSP in the first and second parity, respectively. Heritability estimates for BSO and BSP were low at the beginning of lactation, reached the peak at around eight months in milk, then decrease to the end of lactation (Figure 1). The genetic correlations estimated on a daily basis between BSO and milk traits (MY, FP, and PP) in the first and second parities are presented in Figures 2 and 3, respectively. Mean genetic correlation estimates between BSO and MY, FP, and PP were, respectively, -0.30, -0.38, and -0.01 in the first and -0.31, -0.46 and -0.12 in the second parity. The genetic correlations estimated on a daily basis between BSP and milk traits (MY, FP, and PP) in the first and second parities are presented in Figures 4 and 5, respectively. Mean genetic correlation estimates between BSP and MY, FP, and PP were, respectively, -0.27, -0.38, and -0.06 in the first and -0.27, -0.45 and -0.17 in the second parity. Mean (SD) genetic correlations estimated in daily basis between BSO and BSP were 0.96 (0.01) and 0.95 (0.02), in the first and second parity (Figures 6). Genetic correlations were very high throughout the lactation, indicating that BSO and BSP were two nearly identical traits.

## Conclusions

In this study, two new traits related to milk fatty acid profile associated to technological properties were defined: butter softness (BSO) defined as the ratio of unsaturated to saturated FAs, and butter spreadability (BSP) defined as the ratio of C18:1 (cis-9) FA to C16:0 FA. The heritability estimates of BSO and BSP, as two technological traits, were moderate but varied within lactation. The BSO and BSP showed moderate negative genetic correlations with MY and FP. Soyeurt et al. (2008) reported negative genetic correlation between butter hardness (defined as the ratio of SFA to UFA) and MY, and positive genetic correlation between butter hardness and FP and PP in Holstein cows. The genetic correlations between BSO and BSP were very high, therefore these are nearly identical traits. However, they showed differences in heritability with BSO showing higher values than BSP.

## Implications and Future Developments

Currently, selection in DPBB breed is focusing on increasing MY, FP, and PP. Observed moderate (negative) genetic correlations estimated between the examined traits (BSO and BSP) and milk traits (MY, FP, and PP) indicate that this genetic selection would decrease quality of butterfat. Therefore, research efforts to evaluate genetically BSO and BSP in routine in a joint French-Walloon genomic evaluation and to develop selection indexes restricting negative changes in the future are needed. The ongoing INTERREG VA France-Walloonie-Vlaanderen BlueSter project is supporting these efforts. Very high positive genetic correlation was found between BSO and BSP and this throughout the lactation. However, BSO showed a higher heritability than BSP. In addition, the prediction of larger FA groups like UFA and SFA (used for estimating BSO) is more efficient than individual FA (used for estimating BSP). Furthermore, SFA and UFA

are more frequently available. Therefore, the inclusion of BSO in selection programs is more justified than BSP. Routine genetic evaluations will be developed to be able to benefit from records beyond the second lactation and the large availability of correlated traits (i.e., MY, FP and PP) useful when MIR data to predict BSO and BSP is not yet available as currently in specific regions where DPBB cattle are present. Similarly, MIR based data (i.e., BSO and BSP) will not be available for older records, but, as shown in this study, a specific pattern of correlations across the lactations of MY, FP and PP with BSO and BSP.

Current research can be extended to Holsteins, the major dairy breed in Belgium. In this situation, strategies can be exploited to include external information for INTERBULL evaluated correlated traits using appropriate strategies (Colinet et al., 2018).

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