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# Body and milk production traits as indicators of energy requirements and efficiency of purebred Holstein and 3-breed rotational crossbred cows from Viking Red, Montbéliarde, and Holstein sires

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## ABSTRACT

This study aimed to compare rotational 3-breed crossbred cows of Viking Red, Montbéliarde, and Holstein breeds with purebred Holstein cows for a range of body measurements, as well as different metrics of the cows' productivity and production efficiency. The study involved 791 cows (440 crossbreds and 351 purebreds), that were managed across 2 herds. Within each herd, crossbreds and purebreds were reared and milked together, fed the same diets, and managed as one group. The heart girth, height at withers, and body length were measured, and body condition score (BCS) was determined on all the cows on a single test day. The body weight (BW) of 225 cows were used to develop an equation to predict BW from body size traits, parity, and days in milk, which was then used to estimate the BW of all the cows. Equations from the literature were used to estimate body protein and lipid contents using the predicted BW and BCS. Evidence suggests that maintenance energy requirements may be closely related to body protein mass, and Holstein and crossbred cows may be different in body composition. Therefore, we computed the requirements of net energy for maintenance ($NE_M$) on the basis either of the metabolic weight ($NE_{M\text{-}MW}$: 0.418 MJ/kg of metabolic BW) or of the estimated body protein mass according to a coefficient ($NE_{M\text{-}PM}$: 0.631 MJ/kg body protein mass) computed on the subset comprising the purebred Holstein. On the same day when body measurements were collected, individual test-day milk yield and fat and protein contents were retrieved once from the official Italian milk recording system, and milk was sampled to determine fresh cheese yield. Measures of $NE_M$ were used to scale the production traits. Statistical analyses of all variables included the fixed effects of herd, days in milk, parity, and genetic group (purebred Holstein and crossbred), and the herd × genetic group interaction. External validation of the equation predicting BW yielded a correlation coefficient of 0.94 and an average bias of −4.95 ± 36.81 kg. The crossbreds had similar predicted BW and $NE_{M\text{-}MW}$ compared with the Holsteins. However, $NE_{M\text{-}PM}$ of crossbreds was 3.8% lower than that of the Holsteins, due to their 11% greater BCS and different estimated body composition. The crossbred cows yielded 4.8% less milk and 3.4% less milk energy than the purebred Holsteins. However, the differences between genetic groups were no longer significant when the production traits were scaled on $NE_{M\text{-}PM}$, suggesting that the crossbreds and purebreds have the same productive ability and efficiency per unit of body protein mass. In conclusion, measures of productivity and efficiency that combine the cows' production capability with traits related to body composition and the energy cost of production seem to be more effective criteria for comparing crossbred and purebred Holstein cows than just milk, fat, and protein yields.
**Key words:** crossbreeding, body size, body condition score, milk yield traits, Holstein Friesian

## INTRODUCTION

The Holstein (**HO**) has become the predominant dairy breed due to a huge increase in milk yield resulting from extremely effective selection for production (Hazel et al., 2021; Magne and Quénon, 2021). However, the increase in productivity has been accompanied by a decline in female fertility, health, and longevity, although some improvements in the genetic trends for these traits have been recently reported (Oltenacu and Broom, 2010; Ma et al., 2019; Brito et al., 2021). Therefore, the interest in crossbreeding programs has grown as a means to exploit breed complementarity, reduce inbreeding depression, and capture the benefits

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of heterosis, especially with regard to functional traits (Sørensen et al., 2008; Buckley et al., 2014; Hazel et al., 2020b).

Indeed, rotational 2- or 3-way crossbreeding schemes using a range of modern breeds may exert favorable effects on animal performance by improving the fertility and longevity of dairy herds (Buckley et al., 2014; Malchiodi et al., 2014a) and enhancing profitability (Sørensen et al., 2008; Clasen et al., 2020). Specifically, the 3-breed rotation of the Viking Red (**VR**), Montbéliarde (**MO**), and HO breeds is gaining interest by dairy producers globally (Shonka-Martin et al., 2019a). The VR breed is a combination of different populations of Danish Red, Swedish Red, and Finnish Ayrshire, whose selection index lays particular emphasis on health, disease resistance, and fertility (Viking Genetics, 2023). The MO breed has lower milk yield, greater fertility, and better health traits and milk quality than the HO, and has also been actively selected for improving beef traits (Balandraud et al., 2018). Moreover, fertility and longevity are further improved by heterosis in crossbred cows (**CB**) compared with purebreds (Sørensen et al., 2008).

Nevertheless, many farmers seem reluctant to adopt crossbreeding as a systematic mating program, mainly because purebred HO have higher milk yields than CB (Buckley et al., 2014; Magne and Quénon, 2021). Indeed, several studies reported a greater milk yield for purebred HO than for CB obtained from VR, MO, and HO sires (Malchiodi et al., 2014b; Hazel et al., 2017b; Shonka-Martin et al., 2019a), although higher fat and protein content of milk from CB partly counterbalance the difference in terms of fat plus protein yield (Shonka-Martin et al., 2019a; Saha et al., 2020). Milk, fat, and protein yields are indicators of production, whereas efficiency may be expressed with ratio-based traits, which relate milk outputs to feed inputs or to traits correlated with feed inputs, such as BW and metabolic weight (Connor, 2015; Köck et al., 2018; Berry and McCarthy, 2021). In terms of efficiency of production, differences in BW are of paramount importance when purebred HO are compared with smaller CB, such as those obtained from crossbreeding schemes involving the Jersey breed, often used in low-input, pasture-based systems (Prendiville et al., 2009; Evers et al., 2021).

Research at the University of Minnesota has reported similar BW for HO and CB cows of the 3-breed rotational system involving VR, MO, and HO breeds (Shonka-Martin et al., 2019a). As BW measurements have been used as scaling factors of lactation yield to define efficiency indicators (Macdonald et al., 2008; Lembeye et al., 2016; O'Sullivan et al., 2019), such kinds of metrics are expected to be similar between HO and CB or better in purebred HO. However, the few studies monitoring DMI at the individual cow level seem to indicate that CB are more efficient than HO cows (Shonka-Martin et al., 2019b; Pereira et al., 2022).

We hypothesized that this apparent contradiction could be explained, at least in part, by potential differences in the body compositions of CB and purebred HO. This may lead to overestimation of the maintenance energy requirements of CB when they are based on metabolic weight (**MW**), as equations relating MW to net energy requirements for maintenance were developed using HO (Moraes et al., 2015). Indeed, in using MW, which scales BW by a power ($MW = BW^{0.75}$), it is assumed that cows with similar BW also have similar body compositions, and therefore similar daily $NE_M$ requirements. So, the use of MW for estimating the $NE_M$ requirements of CB and purebred HO rests on the assumption that the cows of both genetic types at the same BW have comparable body composition, and consequently the same proportions of fat, protein, and water. However, several studies have shown that the BCS of CB were greater than those of their purebred HO herdmates (Hazel et al., 2017a; Shonka-Martin et al., 2019a; Hazel et al., 2020b). Differences in BCS reflect differences in body lipid and protein mass (Fox et al., 1999; NRC, 2016; NASEM, 2021), and increasing evidence suggests that maintenance requirements are more closely related to body protein mass than to MW (Agnew and Yan, 2000; Yang et al., 2020; NASEM, 2021). In the case of beef cattle, comparing animals characterized by very different body compositions, such as UK-bred conventional steers and double-muscled young bulls, Schiavon and Bittante (2012) have shown the bias inherent in using MW to predict $NE_M$ and demonstrated the need to consider body composition.

Our objective was to compare CB cows of the 3-breed rotational mating system involving VR, MO, and HO sires with purebred HO for a range of body measurements, BCS, and production metrics in terms of milk, fat, and protein yields and predictors of $NE_M$ based on MW or on predicted body protein mass.

## MATERIALS AND METHODS

Samples and data were collected on commercial dairy farms according to procedures compliant with the Italian legislation concerning the care and safeguard of animals (Italian Legislative Decree n. 26, March 4, 2014).

### *Experimental Design, Herds, and Cows*

The present study involved 791 dairy cows kept on 2 specialized dairy farms (herds A and B, comprising 232 and 559 cows, respectively) located in northern Italy. The single cow was the experimental unit. Both farms produced milk for the production of protected designation of origin hard cheeses (Grana Padano and Parmigiano-Reggiano in herds A and B, respectively). The cows were kept in freestalls with cubicles and were milked twice a day.

The cows of herd A were fed TMR based on corn and sorghum silages and concentrates, according to the guidelines of the Grana Padano cheese consortium (https://www.granapadano.it/en-ww/production-specification-rules.aspx). The cows of herd B were fed TMR based on alfalfa and meadow hay and concentrates and without the use of ensiled forages, in compliance with the regulations of the Parmigiano-Reggiano cheese consortium (https://www.parmigianoreggiano.com/consortium-specifications-and-legislation).

The herds of both dairy farms comprised purebred HO and CB obtained from the 3-breed rotational crossbreeding system known as ProCROSS, according to the mating design described in detail by Saha et al. (2020) and Hazel et al. (2021). Herd A consisted of 147 purebred HO and 85 CB, whereas herd B consisted of 204 purebred HO and 355 CB. Crossbred cows included the following generations: $F_1$ [VR(HO) and MO(HO), n = 139], $F_2$ [VR(MO-HO) and MO(VR-HO), n = 124], and $F_3$ [HO(VR-MO-HO) and HO(MO-VR-HO), n = 177]. Within each herd, purebred HO and CB were reared and milked together, fed the same diets, and managed as one group.

## Milk Yield, Composition, and Cheese-Making Procedure

Milk yield and composition were from one single test day. Individual milk yield and fat and protein contents (MilkoScan FT 6000 infrared analyzer, Foss A/S) were retrieved from the official Italian milk recording system for all 791 cows.

On the same day of the official milk sampling, one further aliquot of milk (50 mL) was collected once during the evening milking from each of the 791 cows. These samples were stored without preservative in a refrigerator at −20°C and transferred to the Milk Laboratory of the University of Padova Department of Agronomy, Food, Natural Resources, Animals and Environment (Legnaro, Italy). The lactose content was measured at the Milk Laboratory with a MilkoScan FT2 infrared analyzer (Foss A/S), and individual curd yields (**$CY_{CURD}$**) were measured using the 9-MilCA method (Cipolat-Gotet et al., 2016) according to a procedure which is comprehensively described for these samples in Saha et al. (2020). Briefly, after heating each 9-mL sample of milk to 35°C, 0.2 mL of 1.2% diluted (wt/vol) rennet solution (Hansen Standard 215 with 80 ± 5% chymosin and 20 ± 5% pepsin; Pacovis Amrein AG) was added, and the temperature was maintained at 35°C for 30 min. After a first manual cut, the samples were heated to 55°C for another 30 min and, in the middle of this cooking phase, were manually cut again. The curd was then separated from the whey for 30 min at room temperature and gently pressed to expel the whey. The resulting curd was weighed using precision scales to determine $CY_{CURD}$, expressed as a percentage of the milk processed.

## Body Trait Measurements and BCS

The heart girth (**HG**, around the cow behind the shoulder), height at withers (**HW**, from the floor to the top of the back in a line up the middle of the shoulder), and body length (**BL**, from front tip of shoulder to edge of pin bone) of all 791 cows in the study were measured once by the same operator on the same day of milk recording. Simultaneously, BCS was assigned to each cow independently of her genetic group by the same skilled operator, according to Edmonson et al. (1989), from 1 (lean) to 5 (fat) in increments of 0.25.

On a subsample of 227 cows from herd B (60 purebred HO and 167 CB), individual BW was also measured using an electronic weighing scale. Body weight was collected on the same day the body traits were measured after the morning milking but before feeding, to develop reliable estimation equations for predicting BW to be applied to all cows in the study.

## Estimation of Body Composition and Net Energy Requirements for Maintenance

The body composition of all cows was estimated using the equations proposed for dairy cows by Fox et al. (1999), cited by NRC (2016). As these equations refer to empty body composition and BCS on a scale of 1 to 9, we modified them to reflect BW (assuming empty BW = 0.82 BW; NASEM, 2021) and BCS on a scale of 1 to 5. The resulting equations were used to estimate the total fat and protein masses of each cow as follows:

$$\text{Body fat mass (kg)} = (0.06171 \times \text{BCS} - 0.0308706) \times \text{BW};$$

$$\text{Body protein mass (kg)} = (-0.01287 \times \text{BCS} + 0.170174) \times \text{BW; and}$$

$$\text{Body water and ash mass (kg)} = (-0.05076 \times \text{BCS} + 0.680697) \times \text{BW}.$$

The total body energy content of each cow was estimated assuming energy values of 38.49 MJ/kg for body fat and 23.22 MJ/kg for body protein (NASEM, 2021).

To account for the differences in body composition between purebred HO and CB, the $NE_M$ of the cows was calculated using 2 different methods. The first one uses the NASEM (2021) equation, in which $NE_M$ is based on MW ($NE_{M\text{-}\mathbf{MW}} = 0.418 \times BW^{0.75}$, MJ/d). The other equation was developed assuming the cow's average daily $NE_M$ to be primarily due to the lean tissues, and then to body protein mass (Agnew and Yan, 2000; NASEM, 2021). Given this, and considering that equations relating MW to net energy requirements for maintenance have been developed on purebred HO (Moraes et al., 2015; NASEM, 2021), we (1) computed the daily $NE_{M\text{-}MW}$ using the NASEM (2021) equation for the group of 351 purebred HO only; (2) computed the coefficient to calculate the $NE_M$ based on protein mass ($NE_{M\text{-}\mathbf{PM}}$) by dividing the $NE_{M\text{-}MW}$ by the body protein mass of each cow of the 351 purebred HO group; and (3) multiplied the average value of this coefficient (0.631 MJ/kg body protein) by the protein mass of each of the 791 cows (purebred HO and CB) to calculate individual predicted $NE_{M\text{-}PM}$ values. Therefore, the second method used the following equation: $NE_{M\text{-}PM} = 0.631 \times$ body protein mass (MJ/d).

## Production Metrics

Based on the traits derived from the milk analysis, we computed the net energy content of the milk according to the following equation (NASEM, 2021):

$$\text{Net energy content (MJ/kg)} = 0.3887 \times \text{fat} + 0.2301 \times \text{protein} + 0.1653 \times \text{lactose},$$

where fat, protein, and lactose are the percentages of fat, protein, and lactose resulting from the official Italian milk recording system (http://bollettino.aia.it/).

Six individual yield indicators—test-day yields of raw milk, fat, protein, fat plus protein, milk energy, and fresh curd—were computed for each cow by multiplying the test-day milk yield by the corresponding traits retrieved from the official Italian milk recording system (test-day fat and protein content) and from the milk analysis and cheesemaking procedures (test-day lactose and $CY_{CURD}$).

Finally, to obtain production efficiency metrics, 12 productivity indicators per cow were computed by scaling the 6 abovementioned test-day yield indicators by the estimated $NE_{M\text{-}MW}$ and $NE_{M\text{-}PM}$.

## Editing and Statistical Analysis

***Body Weight Prediction.*** The preliminary editing step aimed at handling extreme values resulted in a final data set of 225 cows (60 purebred HO and 165 CB), which were grouped into 3 parity (**PAR**) classes (PAR 1, 2, ≥3 with 77, 71, and 77 cows, respectively) and 5 DIM classes of 60 d each (from ≤60 to >240 d with 41 to 53 cows per class). These categorical data were coded as dummy variables (0 or 1). The data set was split into 2 subsets: two-thirds of the cows (n = 150) were used to develop a calibration equation for predicting BW, and one-third (n = 75) of the cows were used to validate the prediction equation. Pearson correlations were computed to assess multicollinearity among traits treated as predictors in subsequent analyses.

A multiple regression model was applied to the calibration data set using the PROC REG function of SAS (version 9.4; SAS Institute Inc.) with a stepwise procedure that included the following independent variables: HG, HW, and BL as continuous variables, and DIM and PAR as dummy variables. The best prediction equation was as follows:

$$\begin{aligned} BW = {} & -700.67 + 18.72 \times PAR_2 + 25.06 \times PAR_3 \\ & + 18.85 \times DIM_2 + 6.98 \times DIM_3 + 9.58 \times DIM_4 \\ & + 15.59 \times DIM_5 + (-3.35 \times HW) \\ & + (6.70 \times HG) + (2.59 \times BL), \end{aligned}$$

where $PAR_2$ and $PAR_3$ were the cows in the second and third parities, respectively, and $DIM_2$, $DIM_3$, $DIM_4$, and $DIM_5$ were the cows of 61 to 120, 121 to 180, 181 to 240, and >240 DIM, respectively.

To test its performance, the equation was used to obtain predicted values for the validation data set and the residuals for evaluation. Regression of the residuals obtained from the equation revealed uniform residual patterns, indicating no bias for either genetic group. Moreover, Bland-Altman plots were created to evaluate agreement between predicted and observed measures (Bland and Altman, 1999). The differences between the predicted and observed BW for each of the 75 cows of the validation data set were computed and used to calculate the average bias and its standard deviation (SD), and subsequently the lower and upper limits of agreement [bias ± (1.96 × SD)]. These parameters have been depicted in a scatter plot, in which the y-axis shows the difference and the x-axis the average of the predicted and observed BW (Figure 1).

Finally, the equation was applied to predict the BW of all 791 cows in the study.

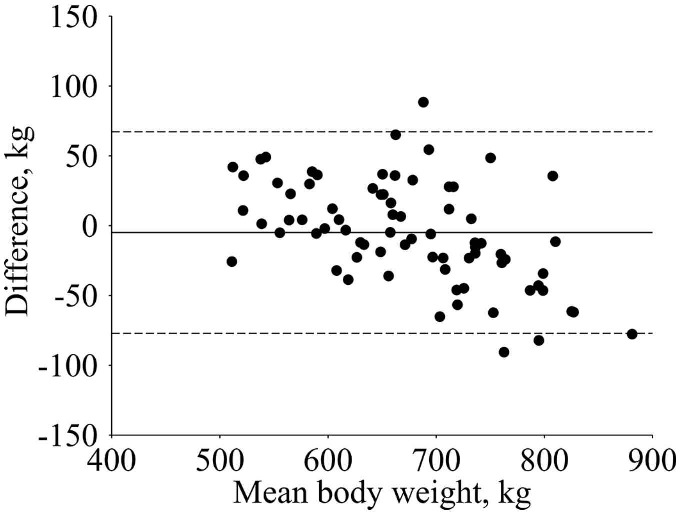

**Figure 1.** Bland-Altman plot illustrating the relationships between BW measured using an electronic weighing scale and BW predicted on an independent validation data set (n = 75 cows) using different body measures (heart girth, height at withers, and body length), parity, and DIM classes. The x-axis is the mean of BW measured and predicted, and the y-axis is the difference between BW predicted and BW measured. The solid line in the middle represents the mean of the difference; the upper and lower dotted lines represent the mean ± 1.96 SD.

***Statistical Analysis.*** All records were classified for PAR (3 classes: 1, 2, and $\geq 3$ with 304, 241, and 246 cows, respectively), DIM (5 classes of 60 d each, from $\leq 60$ to $>240$ d with 127 to 203 cows per class), herd of origin (2 classes), and genetic group (2 classes: purebred HO and CB). Crossbred cows have been taken as a mixture of generations and sire breeds representing the 3-breed rotational system, so comparison of the sire breeds within CB was outside the scope of this study.

After a preliminary exploratory data analysis to identify outliers and the assumptions required for model fitting and hypothesis testing, the milk and body traits and productivity indicators were treated as dependent variables and analyzed using the following linear model in SAS PROC GLM (version 9.4; SAS Institute Inc.):

$$\mathrm{y_{ijklm}} = \mu + \mathrm{DIM_i} + \mathrm{PAR_j} + \mathrm{GG_k} + \mathrm{HD_l} + (\mathrm{GG} \times \mathrm{HD})_{\mathrm{kl}} + \mathrm{e_{ijklm}}.$$

In this model, $\mathrm{y_{ijklm}}$ is the observed trait (i.e., body, milk, and productivity); $\mu$ is the overall mean; $\mathrm{DIM_i}$ is the fixed effect of the ith class of days in milk (i = 5); $\mathrm{PAR_j}$ is the fixed effect of the jth parity (j = 3); $\mathrm{GG_k}$ is the fixed effect of the kth class of the genetic group (k = 2); $\mathrm{HD_l}$ is the fixed effect of the lth herd (l = 2); $(\mathrm{GG} \times \mathrm{HD})_{\mathrm{kl}}$ is the 2-way interaction between $\mathrm{GG_k}$ and $\mathrm{HD_l}$; and $\mathrm{e_{ijklm}}$ is the random residual, assumed to be normally distributed with a mean of zero and a variance of $\sigma_e^2$. A given effect was declared significant at $P < 0.05$ and tendential at $P > 0.05$ but $\leq 0.10$.

## RESULTS

### *Body Weight Prediction*

In the equation predicting BW from body size measurements, PAR, and DIM had a coefficient of determination of 0.81 in calibration and a residual standard error of 36.7 kg. External validation, achieved by regressing the observed and predicted values on an independent validation data set, yielded a coefficient of correlation of 0.94 and an average bias (predicted BW − observed BW) of −4.95 ± 36.81 kg. Bland-Altman plots (Figure 1) included zero within the 95% interval of agreement, and 96% of the difference between the predicted and measured BW was encompassed in the 95% confidence interval of agreement.

### *Descriptive Statistics and the Results of the ANOVA*

Descriptive statistics and the results of the ANOVA for milk composition, $\mathrm{CY_{CURD}}$, body traits, and $\mathrm{NE_M}$ are given in Table 1. The average percentage contents of fat and protein were 3.75 and 3.64%, respectively, and the variation for fat content was nearly twice the variation for protein content. Average body size measures were HW 140 cm, BL 164 cm, and HG 209 cm, which gave an average predicted BW of about 680 kg.

As expected, herd, PAR, and DIM had significant effects on the vast majority of traits. Genetic group significantly influenced the milk protein content ($P < 0.01$), the $\mathrm{CY_{CURD}}$ ($P < 0.05$), the BCS ($P < 0.01$), and all body size measurements but not the predicted BW. Also predicted body composition was significantly different in the 2 genetic groups ($P < 0.01$), whereas $\mathrm{NE_M}$ was similar when computed from MW but significantly different when computed from body protein mass. Finally, the effect of the herd × genetic group interaction was never significant.

Descriptive statistics and the results of the ANOVA for the production metrics are given in Table 2. In this study, the cows yielded on average nearly 33.9 kg/d of milk, equivalent to 5.7 kg/d of fresh curd and 2.5 kg/d of fat plus protein, which, when scaled to $\mathrm{NE_M}$ based on either MW or protein mass, were around 610 to 620 g/MJ, 102 to 104 g/MJ, and 44 to 45 g/MJ, respectively. The coefficient of variation ranged from 25 to 30% for all production metrics.

Again, herd, PAR, and DIM significantly affected all of the various production traits ($P < 0.01$). Genetic

**Table 1.** Raw means, SD, and *P*-values from ANOVA for milk composition, fresh curd yield, body traits, and estimated NE$_{\text{M}}$ requirements based on metabolic weight (NE$_{\text{M-MW}}$) and on body protein mass (NE$_{\text{M-PM}}$)[1]

| | | | *P*-value[2] | | | | | |
|---|---|---|---|---|---|---|---|---|
| Trait | Raw mean | SD | Herd | Parity | DIM | GG | Herd × GG | RMSE |
| Milk composition and cheese yield | | | | | | | | |
| Fat content, % | 3.75 | 0.75 | 0.006 | 0.10 | <0.001 | 0.24 | 0.78 | 0.72 |
| Protein content, % | 3.64 | 0.35 | <0.001 | 0.07 | <0.001 | 0.004 | 0.14 | 0.29 |
| Energy content, MJ/kg | 3.13 | 0.34 | 0.12 | 0.01 | <0.001 | 0.13 | 0.51 | 0.32 |
| Fresh curd yield, % | 16.84 | 2.25 | 0.02 | 0.54 | <0.001 | 0.045 | 0.73 | 2.20 |
| Body trait | | | | | | | | |
| BCS (1 to 5 score) | 3.26 | 0.37 | <0.001 | <0.001 | <0.001 | <0.001 | 0.06 | 0.32 |
| Body length, cm | 164 | 7 | <0.001 | <0.001 | <0.001 | <0.001 | 0.22 | 6 |
| Heart girth, cm | 209 | 9 | <0.001 | <0.001 | <0.001 | 0.004 | 0.63 | 8 |
| Height at withers, cm | 140 | 5 | <0.001 | <0.001 | 0.01 | <0.001 | 0.25 | 4 |
| Predicted BW, kg | 677 | 72 | <0.001 | <0.001 | <0.001 | 0.30 | 0.69 | 54 |
| Body composition[3] | | | | | | | | |
| Predicted body fat mass, kg | 116 | 23 | <0.001 | <0.001 | <0.001 | <0.001 | 0.16 | 20 |
| Predicted body protein mass, kg | 87 | 9 | 0.02 | <0.001 | <0.001 | <0.001 | 0.82 | 6 |
| Predicted body energy content, MJ | 6,492 | 1,003 | <0.001 | <0.001 | <0.001 | <0.001 | 0.21 | 866 |
| Maintenance requirement[4] | | | | | | | | |
| NE$_{\text{M-MW}}$, MJ/d | 55.39 | 4.42 | <0.001 | <0.001 | <0.001 | 0.31 | 0.55 | 3.31 |
| NE$_{\text{M-PM}}$, MJ/d | 54.68 | 5.50 | 0.02 | <0.001 | <0.001 | <0.001 | 0.82 | 3.68 |

[1] n = 791 cows comprised of 351 purebred Holstein and 440 crossbreds.

[2] Parity = first, second, and third and later lactations; DIM = days in milk classes: ≤60, 61 to 120, 121 to 180, 181 to 240, and ≥241 d; GG = genetic group (Holstein and crossbred cows); RMSE = root mean square error.

[3] Predicted body fat mass (kg) = (0.06171 × BCS − 0.0308706) × BW; predicted body protein mass (kg) = (−0.01287 × BCS + 0.170174) × BW; predicted body energy content (MJ) = 23.22 × predicted body protein mass + 38.49 × predicted body fat mass, according to NRC (2016) and NASEM (2021).

[4] NE$_{\text{M-MW}}$ (MJ/d) = 0.418 × BW$^{0.75}$; NE$_{\text{M-PM}}$ (MJ/d) = 0.631 × predicted body protein mass.

**Table 2.** Raw means, standard deviation, and *P*-values from ANOVA for yield of milk, milk components, milk energy, and fresh curd in absolute value (daily yield) or scaled on NE$_{\text{M}}$ requirements based on metabolic weight (NE$_{\text{M-MW}}$) and on body protein mass (NE$_{\text{M-PM}}$)[1]

| | | | *P*-value[2] | | | | | |
|---|---|---|---|---|---|---|---|---|
| Trait | Raw mean | SD | Herd | Parity | DIM | GG | Herd × GG | RMSE |
| Milk production | | | | | | | | |
| Daily yield, kg/d | 33.85 | 10.11 | <0.001 | <0.001 | <0.001 | 0.01 | 0.49 | 7.74 |
| Daily yield/NE$_{\text{M-MW}}$, g/MJ | 613 | 182 | <0.001 | <0.001 | <0.001 | 0.02 | 0.66 | 147 |
| Daily yield/NE$_{\text{M-PM}}$, g/MJ | 621 | 178 | <0.001 | <0.001 | <0.001 | 0.58 | 0.44 | 146 |
| Fat production | | | | | | | | |
| Daily yield, g/d | 1,240 | 371 | <0.001 | <0.001 | <0.001 | 0.14 | 0.31 | 306 |
| Daily yield/NE$_{\text{M-MW}}$, g/MJ | 22.5 | 6.6 | <0.001 | 0.001 | <0.001 | 0.21 | 0.48 | 5.7 |
| Daily yield/NE$_{\text{M-PM}}$, g/MJ | 22.8 | 6.6 | <0.001 | 0.04 | <0.001 | 0.63 | 0.29 | 5.7 |
| Protein production | | | | | | | | |
| Daily yield, g/d | 1,214 | 316 | <0.001 | <0.001 | <0.001 | 0.08 | 0.11 | 258 |
| Daily yield/NE$_{\text{M-MW}}$, g/MJ | 22.0 | 5.6 | 0.003 | <0.001 | <0.001 | 0.15 | 0.19 | 4.9 |
| Daily yield/NE$_{\text{M-PM}}$, g/MJ | 22.2 | 5.5 | <0.001 | <0.001 | <0.001 | 0.61 | 0.10 | 4.9 |
| Fat plus protein production | | | | | | | | |
| Daily yield, g/d | 2,454 | 647 | <0.001 | <0.001 | <0.001 | 0.08 | 0.16 | 521 |
| Daily yield/NE$_{\text{M-MW}}$, g/MJ | 44.4 | 11.5 | <0.001 | <0.001 | <0.001 | 0.15 | 0.29 | 9.8 |
| Daily yield/NE$_{\text{M-PM}}$, g/MJ | 45.0 | 11.4 | <0.001 | <0.001 | <0.001 | 0.59 | 0.15 | 9.8 |
| Milk energy production | | | | | | | | |
| Daily yield, MJ/d | 104.6 | 28.4 | <0.001 | <0.001 | <0.001 | 0.04 | 0.22 | 22.2 |
| Daily yield/NE$_{\text{M-MW}}$, MJ/MJ | 1.89 | 0.51 | <0.001 | <0.001 | <0.001 | 0.08 | 0.37 | 0.42 |
| Daily yield/NE$_{\text{M-PM}}$, MJ/MJ | 1.92 | 0.50 | <0.001 | 0.003 | <0.001 | 0.85 | 0.20 | 0.42 |
| Fresh curd production | | | | | | | | |
| Daily yield, kg/d | 5.65 | 1.70 | <0.001 | <0.001 | <0.001 | 0.14 | 0.52 | 1.41 |
| Daily yield/NE$_{\text{M-MW}}$, g/MJ | 102 | 30 | <0.001 | <0.001 | <0.001 | 0.22 | 0.65 | 26 |
| Daily yield/NE$_{\text{M-PM}}$, g/MJ | 104 | 30 | <0.001 | 0.01 | <0.001 | 0.64 | 0.45 | 26 |

[1] n = 791 cows, comprising 351 purebred Holstein and 440 crossbreds. NE$_{\text{M-MW}}$ (MJ/d) = 0.418 × BW$^{0.75}$; NE$_{\text{M-PM}}$ (MJ/d) = 0.631 × predicted body protein mass. Predicted body protein mass (kg) = (−0.01287 × BCS + 0.170174) × BW.

[2] Parity = first, second, and third and later lactation; DIM = days in milk classes: ≤60, 61 to 120; 121 to 180; 181 to 240; ≥241 d; GG = genetic group (Holstein and crossbred cows); RMSE = root mean square error.

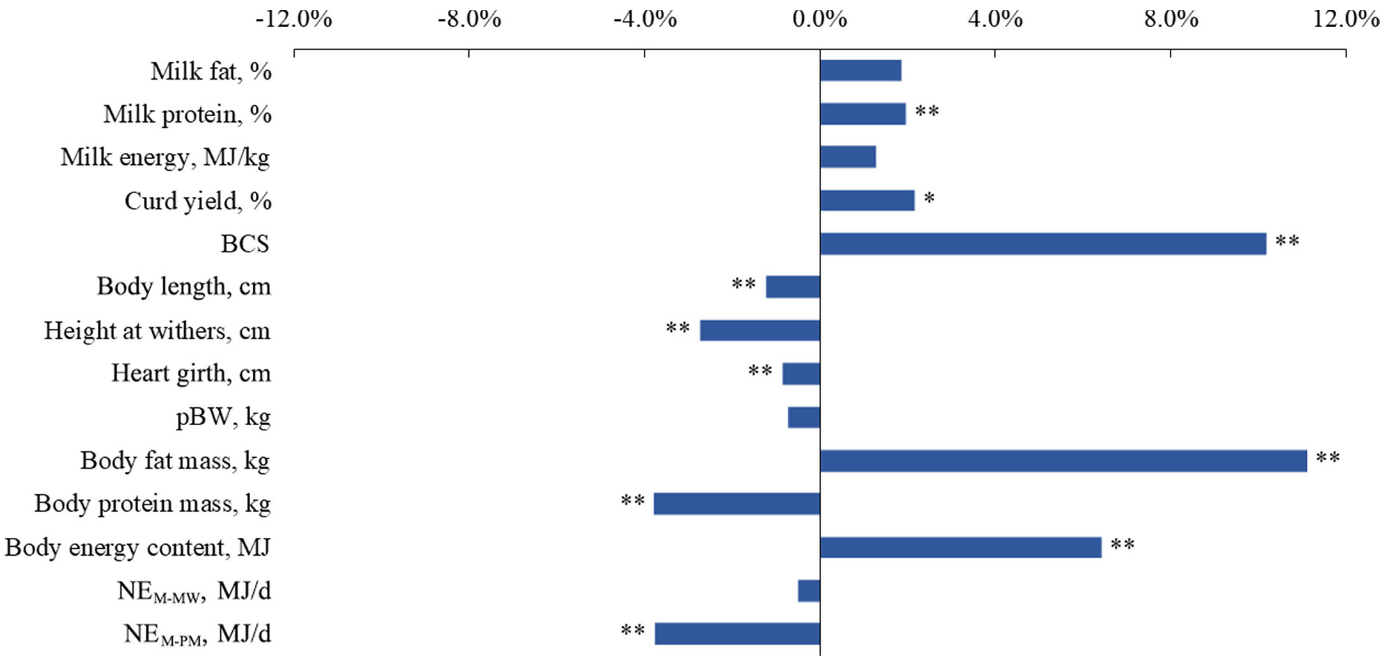

**Figure 2.** Percentage deviation of least squares means of crossbred cows with respect to least squares means of Holstein cows for milk fat, protein, and energy content; curd yield; BCS; body measures; predicted body weight (pBW); body fat, protein, and energy content; and NE$_M$ requirements based on metabolic weight (NE$_{M-MW}$) or on body protein mass (NE$_{M-PM}$). Asterisks refer to the level of significance of differences between crossbred and purebred Holstein cows (**$P < 0.01$; *$P < 0.05$).

group significantly influenced the yield of milk volume and milk energy, whereas the yield of protein and fat plus protein was only tendentially different in the 2 genetic groups ($P = 0.08$). When the yield traits were scaled on NE$_{M-MW}$, differences between genetic groups concerned only raw milk ($P < 0.05$) and, tendentially, milk energy ($P = 0.08$). When the yields of output were scaled on NE$_{M-MW}$, genetic group did not influence any production trait. No significant herd × genetic group interaction was detected.

### Crossbred and Purebred Cow Comparisons

The protein content and the CY$_{CURD}$ were nearly 2% greater in the milk produced by the CB compared with the purebred HO (Figure 2).

Concerning the biometric measures, CB had 1.2, 2.7, and 0.9% shorter BL, HW, and HG, respectively, than purebred HO, where, as predicted, BW was similar in the 2 groups of cows. The average BCS of the CB was 10.2% higher than that of the purebred HO, which resulted in the CB having ($P < 0.01$) 11% greater estimated body fat mass, 6.4% greater estimated body energy content, and 3.8% lower estimated body protein mass compared with the purebred HO (Figure 2).

Maintenance energy requirements were similar for the CB and purebred HO when computed from MW but 3.8% lower for CB when computed from body protein mass.

The milk yield of the CB was 4.8% lower than that of the purebred HO when expressed as raw milk (Figure 3) and 3.4% lower when the output was measured as milk energy, whereas fat plus protein yield was only tendentially lower (−2.9%, $P = 0.08$) in CB compared with purebred HO.

The differences between CB and purebred HO slightly decreased when the yield traits were scaled on NE$_{M-MW}$ but remained significant for raw milk (−4.4%, $P < 0.05$) and tendential for milk energy (−3%, $P = 0.08$). When the yields of outputs were scaled on NE$_{M-PM}$, the differences between the 2 genetic groups ranged from −1 to +1%, according to the production metric, with no statistical difference between groups.

## DISCUSSION

### Body Weight Prediction

In the current study, the best equation for estimating BW was obtained using HG, HW, and BL measurements as independent variables. As BW changes with age across lactations and with DIM within lactation, such sources of variation have been included in the equation proposed. When the measure of individual

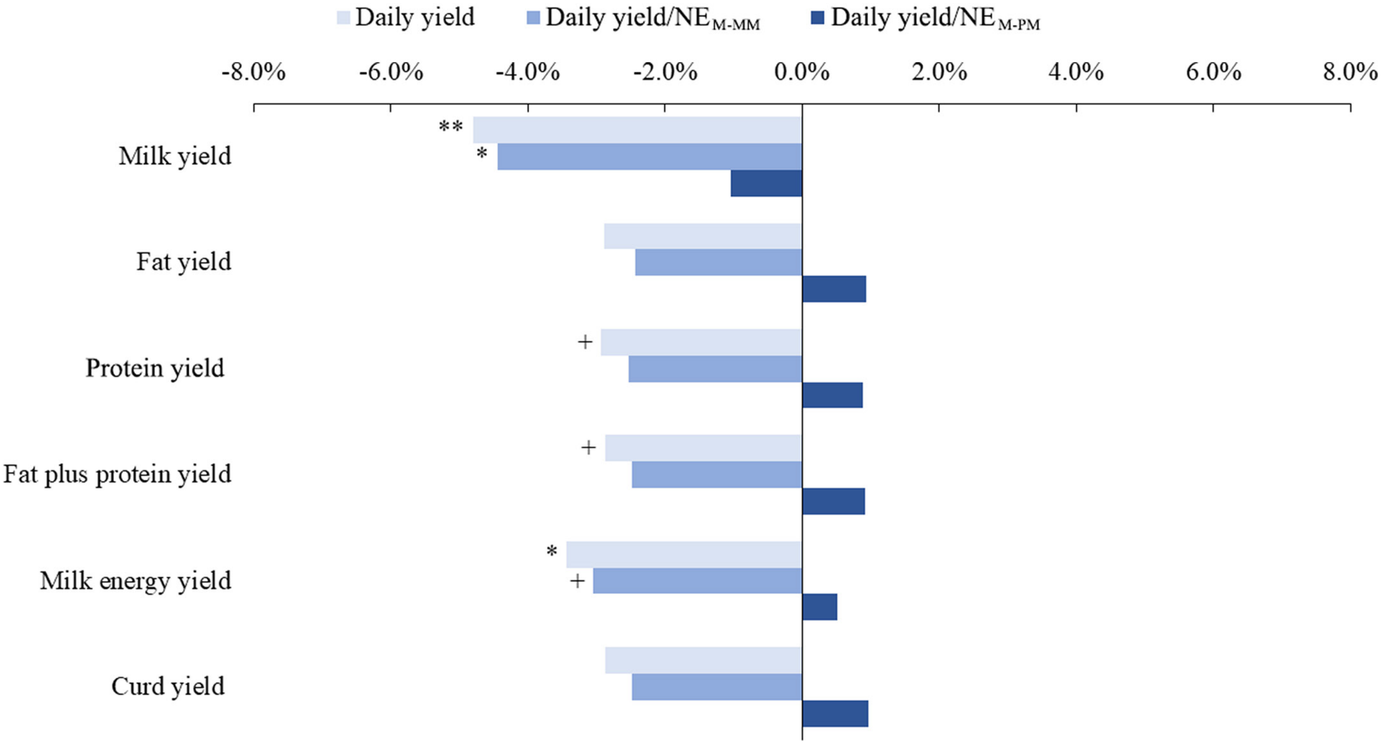

**Figure 3.** Percentage deviation of least squares means of crossbred cows with respect to least squares means of Holstein cows for daily yield of milk (kg), milk fat and protein (g), milk energy (MJ), and fresh curd (g) in absolute value or scaled on NE$_M$ requirements based on metabolic weight (NE$_{M-MW}$, MJ) or on body protein mass (NE$_{M-PM}$, MJ). Symbols refer to the level of significance of differences between crossbred and purebred HO cows (**$P < 0.01$; *$P < 0.05$; +$P < 0.10$).

DMI is unfeasible, the cow's BW has been proposed as a scaling factor to estimate feed efficiency (Berry and McCarthy, 2021). As BW is rarely available, due to the cost of weighing scales and the time required to weigh the animals (Heinrichs et al., 2017), body conformation measures can provide useful predictions of BW in the absence of weighing scales.

Variance inflation factors were <2.5 for all parameters retained in the equation, and we can therefore assume that the traits treated as predictors did not exhibit any appreciable multicollinearity (Johnston et al., 2018). In addition, the Bland-Altman plot evidenced good agreement between the predicted and measured BW, as 96% of the data points lay within ±1.96 SD and the magnitude of average bias was just 0.39% of the average BW (Bland and Altman, 1999).

The performance of the proposed equation is comparable to those of other equations obtained in previous studies: R$^2$ in validation, equal to 0.81 in this study, is nearly identical to that reported by Piazza et al. (2022) in purebred HO cows (0.80), whereas Heinrichs et al. (1992) reported an R$^2$ in calibration above 0.9 using various body measurements to predict the BW of HO heifers. Piazza et al. (2022) found a coefficient of correlation between the observed and predicted BW of 0.88, compared with a coefficient of 0.94 in the present research, whereas the average bias in that study was equal to 8.1 ± 42.9 kg, compared with 4.95 ± 36.8 kg found in this study.

It is worth noting that the BW of an individual cow can change considerably over a short space of time (within a day or days), mainly due to the effects of eating, drinking, defecating, urinating, and milking. Clearly, the predictors used for estimating BW mainly reflect the cow's skeletal development, and, in the case of HG, the cow's fatness and muscularity. As these factors do not change appreciably during the day or from one day to another, we expect the predicted BW to have good repeatability, even though this expectation needs to be confirmed by specific results.

## Body Measures, Body Condition, and Estimated Body Protein and Lipid Masses

The results of the present research evidenced that CB have smaller BL, HG, and HW than HO herdmates but comparable predicted BW. This is due to the different sign of the regression coefficients of the 3 measures on

BW (positive for HG and BL, negative for HW). In addition, Hazel et al. (2017a) found a lower stature score (−19%, −15%, and −14% in first, second, and third lactation, respectively) for pooled MO-HO and VR-HO cows compared with purebred HO. Similar findings have been reported by Hazel et al. (2020b) for 3 generations of CB cows of the same crossbreeding scheme, which scored nearly 20% lower for stature compared with their purebred HO herdmates. Similarly, Shonka-Martin et al. (2019a) found that rotational CB cows of the VR, MO, and HO breeds had 3 and 2.5% shorter HW in their first and later lactations, respectively, than purebred HO but comparable measured BW. In addition, Pereira et al. (2022) recently reported that BW did not differ for CB and purebred HO fed a traditional TMR diet.

Moreover, CB had higher BCS than purebred HO (+10%, equal to 0.32 units of BCS). These results agree with the findings of Hazel et al. (2017a), who found a 10 to 12% greater BCS in MO-HO and VR-HO cows of different parity orders compared with purebred HO, and with the findings of Hazel et al. (2020b), who reported that 3 generations of CB cows of the same crossbreeding scheme evidenced in first lactation a 16% greater BCS compared with first-parity purebred HO. In addition, Shonka-Martin et al. (2019a) found that primiparous and multiparous CB cows of the VR, MO, and HO breeds had greater BCS (+0.26 and +0.19 units, respectively) than purebred HO. Body condition scoring is a rapid, noninvasive, low-cost, subjective method for estimating the cows' degree of fatness (Waltner et al., 1994) and is a valuable tool for dairy farmers to monitor fat and energy changes in the cows during lactation (Edmonson et al., 1989; Gallo et al., 1996). The higher BCS has been called upon to explain some of the CB advantages in fertility over purebred HO (Hazel et al., 2017a). Indeed, decreased reproductive performances have been related to low BCS at first insemination (Bewley and Schutz, 2008), and BCS in early lactation has been negatively associated with days to first estrus and positively associated with an increased likelihood of early estrus (Roche et al., 2009).

In this study, we used the predicted BW and BCS to estimate the body fat and protein masses of cows using equations developed on mature cows of different breed types (NRC, 1996) and modified by Fox et al. (1999) to extend their use to dairy cattle. The average values of body protein masses predicted (21% of empty BW, data not shown in tables) were comparable to the 18 to 19% protein concentration of empty BW measured by Andrew et al. (1994) and Agnew et al. (2005), respectively, in HO cows. Conversely, the empty BW fat content reported by those authors (10 to 17%, according to the stage of lactation) was slightly lower than the average value of fat masses predicted in our study (15.7%).

Despite a similar predicted BW, the CB had higher BCS scores and were therefore estimated to have less body protein, greater body fat mass, and greater body energy content than the purebred HO. To the best of our knowledge, no study in the literature has compared the body composition of CB and purebred HO; however, Piazza et al. (2023) recently reported that carcasses from culled CB of the same crossbreeding scheme were graded 13% higher for fatness compared with culled purebred HO of the same herd.

It should be noted that the precision and the accuracy of equations predicting the body fat and protein masses from body traits and BCS have not been widely studied, mainly because of the scarcity of body composition data, and should therefore be investigated further. Several factors can contribute to the uncertainty of estimates obtained, such as the subjective nature of the method, differences between breeds and individuals in the presence and distribution of fat depots and muscles, and differences in physiological stages and in the proportion of the BW attributable to digesta and milk (Gibb and Ivings, 1993; Gregory et al., 1998). Because of this uncertainty, predictions obtained using this method should be treated with caution and used mainly for comparative purposes under the same productive circumstances, as in the present study.

### Energy Requirement for Maintenance and Production Metrics

In the present study, the CB yielded a significantly lower amount of milk volume than the purebred HO. Looking at the milk components, CB still yielded significantly lower daily milk energy and tended to also have lower fat plus protein yield. These results are in general agreement with findings from the literature, particularly with regard to the volume of milk yielded, which has been consistently found to be lower, in the range of −2 to −10%, in CB of this crossbreeding scheme compared with their purebred HO herdmates (Heins and Hansen, 2012; Malchiodi et al., 2014b; Hazel et al., 2020a). Conversely, the fat plus protein yield of CB was found to be comparable in some studies (Hazel et al., 2014; Shonka-Martin et al., 2019a; Pereira et al., 2022) but greater in purebred HO in others (Heins and Hansen, 2012; Hazel et al., 2020a).

Maximizing milk yield allows dairy operations to dilute maintenance expenses over more units of milk, thus increasing their profitability (VandeHaar and St-Pierre, 2006). As a consequence, the yields of milk and milk solids have become typical metrics for comparing the production potential of dairy cows. However, a possible decline of the dilution of maintenance with further increases in milk yield (Bach et al., 2020) and pressing demands from society to reduce the negative effects on the environment of dairy production has gradually shifted the focus to production efficiency (Berry and Crowley, 2012). Maintenance expenditures account for a substantial proportion of the energy costs in dairy cows and are considered related in most energy systems to the unit of MW (NASEM, 2021). Therefore, milk yield per kilogram of cow BW or MW has been used as a metric of gross efficiency in several studies (Prendiville et al., 2009; Köck et al., 2018; Berry and McCarthy, 2021).

Results of this study evidenced that gross feed efficiency, when expressed by scaling yield traits on $NE_{M\text{-}MW}$, was tendentially or nominally lower in CB than in purebred HO because the 2 genetic groups had similar predicted BW and, consequently, MW. These results are not consistent with findings of Shonka-Martin et al. (2019b), who observed a greater overall feed efficiency for rotational 3-breed CB of this mating scheme compared with purebred HO due to greater fat plus protein yields per unit of DMI in their first 150 d of lactation.

However, the adequacy of MW as the scaling factor for computing $NE_M$ may be questionable, particularly when comparing cows of different breeds, possibly characterized by different body composition. Indeed, relating MW to $NE_M$ presumes that cows of the same BW are expected to have the same body composition and, consequently, the same maintenance requirements, whereas these traits could instead be different in cows of different breeds, yield potentials, body conditions, or other factors. Indeed, Oldham and Emmans (1990) found it physiologically unreasonable to assume $NE_M$ related to MW, given that body composition varies in terms of protein and fat content, and the energy required to maintain 1 kg of protein is much greater than the energy required to maintain 1 kg of fat (DiCostanzo et al., 1990).

Based on these premises, results from this study evidenced that CB had lower $NE_M$ when maintenance requirements have been related to body protein mass. Namely, daily $NE_{M\text{-}PM}$ of CB would be about 2 MJ/d lower than that of the purebred HO (−3.9%). Shonka-Martin et al. (2019b) found lower DMI per unit of BW in CB of the same rotational 3-breed mating scheme and hypothesized that this difference could be due to an enhanced ability of CB to extract nutrients from the diets. Results from this study suggest that such a difference could be explained, at least partially, by also considering possible differences in maintenance energy requirements of CB and purebred HO due to different amounts of body muscle and fat.

When yield traits have been scaled on $NE_{M\text{-}PM}$, all of the differences between the CB and the purebred HO disappeared, and instead, the CB evidenced a slight, although nonsignificant, superiority in terms of fat plus protein and fresh curd yielded per MJ of $NE_{M\text{-}PM}$. Therefore, when $NE_M$ was computed considering the differences in body composition between different genetic groups, the CB showed the same gross feed efficiency, expressed in terms of yield traits per unit of $NE_M$, as purebred HO.

### *Implications for Research and the Industry*

From a scientific point of view, results from this study brought into question the adequacy of MW as the scaling factor in the estimation of the requirements of $NE_M$ for cows having similar BW but different body composition, such as CB and purebred HO. In the case of beef breeds, the inadequacy of MW to provide a correct prediction of $NE_M$ requirements has been mitigated by including correction factors specific for different breeds and breed combinations (NRC, 2016). Clearly, the use of empiric correction factors seems more to represent a rough adjustment rather than a scientific solution. Conversely, the use of body mass composition could explain a large part of the variation observed in both beef and dairy cattle breeds and crosses.

Further research is needed to develop more precise and accurate methods for estimating the body composition of the cows and for evaluating the relationships between body protein mass and the energy expenditure for maintenance. In the meantime, the use of BCS for predicting a proxy of body composition and of $NE_M$ requirements could allow researchers to better understand and quantify the genetic and phenotypic causes of variation of metabolism and the efficiency of dairy cows. Moreover, the industry could benefit from more correct dietary formulations, cost quantification, and profitability comparisons of different genetic groups, including CB.

## CONCLUSIONS

The results of this study showed that CB had similar BW but greater BCS and lower estimated body protein mass compared with their purebred HO herdmates, suggesting that CB could also have lower energy requirements for maintenance. Crossbred cows tended to have lower daily yields of milk, milk energy, and fat plus protein. However, when production metrics were scaled on estimations of energy requirements for maintenance considering differences in body composition, the production performances of CB and purebred HO were entirely comparable. Therefore, measures of efficiency that combine the cows' production ability with traits related to the cost of production seem to be more effective parameters than simply milk and milk solids yields for comparing the outcomes of the crossbreeding mating scheme studied here against purebred HO.

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## REFERENCES

Agnew, R. E., and T. Yan. 2000. Impact of recent research on energy feeding systems for dairy cattle. Livest. Prod. Sci. 66:197–215. https://doi.org/10.1016/S0301-6226(00)00161-5.

Agnew, R. E., T. Yan, W. J. McCaughey, J. D. McEvoy, D. C. Patterson, M. G. Porter, and R. W. J. Steen. 2005. Relationships between urea dilution measurements and body weight and composition of lactating dairy cows. J. Dairy Sci. 88:2476–2486. https://doi.org/10.3168/jds.S0022-0302(05)72925-8.

Andrew, S. M., D. R. Waldo, and R. A. Erdman. 1994. Direct analysis of body composition of dairy cows at three physiological stages. J. Dairy Sci. 77:3022–3033. https://doi.org/10.3168/jds.S0022-0302(94)77244-1.

Bach, A., M. Terré, and M. Vidal. 2020. Symposium review: Decomposing efficiency of milk production and maximizing profit. J. Dairy Sci. 103:5709–5725. https://doi.org/10.3168/jds.2019-17304.

Balandraud, N., C. Mosnier, L. Delaby, F. Dubief, J.-P. Goron, B. Martin, D. Pomiès, and A. Cassard. 2018. Holstein ou Montbéliarde: Des differences phénotypiques aux consequences économiques à l'échelle de l'exploitation. INRA Prod. Anim. 31:337–352. https://doi.org/10.20870/productions-animales.2018.31.4.2394.

Berry, D. P., and J. J. Crowley. 2012. Residual intake and body weight gain: A new measure of efficiency in growing cattle. J. Anim. Sci. 90:109–115. https://doi.org/10.2527/jas.2011-4245.

Berry, D. P., and J. McCarthy. 2021. Contribution of genetic variability to phenotypic differences in on-farm efficiency metrics of dairy cows based on body weight and milk solids yield. J. Dairy Sci. 104:12693–12702. https://doi.org/10.3168/jds.2021-20542.

Bewley, J. M., and M. M. Schutz. 2008. An interdisciplinary review of body condition scoring for dairy cattle. Prof. Anim. Sci. 24:507–529. https://doi.org/10.15232/S1080-7446(15)30901-3.

Bland, J. M., and D. G. Altman. 1999. Measuring agreement in method comparison studies. Stat. Methods Med. Res. 8:135–160. https://doi.org/10.1177/096228029900800204.

Brito, L. F., N. Bédère, F. Douhard, H. R. Oliveira, M. Arnal, F. Peñagaricano, A. P. Schinckel, C. F. Baes, and F. Miglior. 2021. Review: Genetic selection of high-yielding dairy cattle toward sustainable farming systems in a rapidly changing world. Animal 15:100292. https://doi.org/10.1016/j.animal.2021.100292.

Buckley, F., N. Lopez-Villalobos, and B. J. Heins. 2014. Crossbreeding: Implications for dairy cow fertility and survival. Animal 8:122–133. https://doi.org/10.1017/S1751731114000901.

Cipolat-Gotet, C., A. Cecchinato, G. Stocco, and G. Bittante. 2016. The 9-MilCA method as a rapid, partly automated protocol for simultaneously recording milk coagulation, curd firming, syneresis, cheese yield, and curd nutrients recovery or whey loss. J. Dairy Sci. 99:1065–1082. https://doi.org/10.3168/jds.2015-9734.

Clasen, J. B., W. F. Fikse, M. Kargo, L. Rydhmer, E. Strandberg, and S. Østergaard. 2020. Economic consequences of dairy crossbreeding in conventional and organic herds in Sweden. J. Dairy Sci. 103:514–528. https://doi.org/10.3168/jds.2019-16958.

Connor, E. E. 2015. Invited review: Improving feed efficiency in dairy production: Challenges and possibilities. Animal 9:395–408. https://doi.org/10.1017/S1751731114002997.

DiCostanzo, A., J. C. Meiske, S. D. Plegge, T. M. Peters, and R. D. Goodrich. 1990. Within-herd variation in energy utilization for maintenance and gain in beef cows. J. Anim. Sci. 68:2156–2165. https://doi.org/10.2527/1990.6872156x.

Edmonson, A. J., I. J. Lean, L. D. Weaver, T. Farver, and G. Webster. 1989. A body condition scoring chart for Holstein dairy cows. J. Dairy Sci. 72:68–78. https://doi.org/10.3168/jds.S0022-0302(89)79081-0.

Evers, S. H., S. McParland, L. Delaby, K. M. Pierce, and B. Horan. 2021. Analysis of milk solids production and mid-lactation bodyweight to evaluate cow production efficiency on commercial dairy farms. Livest. Sci. 252:104691. https://doi.org/10.1016/j.livsci.2021.104691.

Fox, D. G., M. E. Van Amburgh, and T. P. Tylutki. 1999. Predicting requirements for growth, maturity, and body reserves in dairy cattle. J. Dairy Sci. 82:1968–1977. https://doi.org/10.3168/jds.S0022-0302(99)75433-0.

Gallo, L., P. Carnier, M. Cassandro, R. Mantovani, L. Bailoni, B. Contiero, and G. Bittante. 1996. Change in body condition score of Holstein cows as affected by parity and mature equivalent milk yield. J. Dairy Sci. 79:1009–1015. https://doi.org/10.3168/jds.S0022-0302(96)76452-4.

Gibb, M. J., and W. E. Ivings. 1993. A note on the estimation of body fat, protein and energy content of lactating Holstein-Friesian cows by measurement of condition score and live weight. Anim. Sci. 56:281–283. https://doi.org/10.1017/S0003356100021383.

Gregory, N. G., J. K. Robins, D. G. Thomas, and R. W. Purchas. 1998. Relationship between body condition score and body composition in dairy cows. N. Z. J. Agric. Res. 41:527–532. https://doi.org/10.1080/00288233.1998.9513335.

Hazel, A. R., B. J. Heins, and L. B. Hansen. 2017a. Fertility, survival, and conformation of Montbéliarde × Holstein and Viking Red × Holstein crossbred cows compared with pure Holstein cows during first lactation in 8 commercial dairy herds. J. Dairy Sci. 100:9447–9458. https://doi.org/10.3168/jds.2017-12824.

Hazel, A. R., B. J. Heins, and L. B. Hansen. 2017b. Production and calving traits of Montbéliarde × Holstein and Viking Red × Holstein cows compared with pure Holstein cows during first lactation in 8 commercial dairy herds. J. Dairy Sci. 100:4139–4149. https://doi.org/10.3168/jds.2016-11860.

Hazel, A. R., B. J. Heins, and L. B. Hansen. 2020a. Fertility and 305-day production of Viking Red-, Montbéliarde-, and Holstein-sired crossbred cows compared with Holstein cows during their first 3 lactations in Minnesota dairy herds. J. Dairy Sci. 103:8683–8697. https://doi.org/10.3168/jds.2020-18196.

Hazel, A. R., B. J. Heins, and L. B. Hansen. 2020b. Health treatment cost, stillbirth, survival, and conformation of Viking Red-, Montbéliarde-, and Holstein-sired crossbred cows compared with pure Holstein cows during their first 3 lactation. J. Dairy Sci. 103:10917–10939. https://doi.org/10.3168/jds.2020-18604.

Hazel, A. R., B. J. Heins, and L. B. Hansen. 2021. Herd life, lifetime production, and profitability of Viking Red-sired and Montbéliarde-sired crossbred cows compared with their Holstein herdmates. J. Dairy Sci. 104:3261–3277. https://doi.org/10.3168/jds.2020-19137.

Hazel, A. R., B. J. Heins, A. J. Seykora, and L. B. Hansen. 2014. Production, fertility, survival, and body measurements of Montbéliarde-sired crossbreds compared with pure Holsteins during their first 5 lactations. J. Dairy Sci. 97:2512–2525. https://doi.org/10.3168/jds.2013-7063.

Heinrichs, A. J., B. S. Heinrichs, C. M. Jones, P. S. Erickson, K. F. Kalscheur, T. D. Nennich, B. J. Heins, and F. C. Cardoso. 2017. Short communication: Verifying Holstein heifer heart girth to body weight prediction equations. J. Dairy Sci. 100:8451–8454. https://doi.org/10.3168/jds.2016-12496.

Heinrichs, A. J., G. W. Rogers, and J. B. Cooper. 1992. Predicting body weight and wither height in Holstein heifers using body measurements. J. Dairy Sci. 75:3576–3581. https://doi.org/10.3168/jds.S0022-0302(92)78134-X.

Heins, B. J., and L. B. Hansen. 2012. Short communication: Fertility, somatic cell score, and production of Normande × Holstein, Montbéliarde × Holstein, and Scandinavian Red × Holstein crossbreds versus pure Holsteins during their first 5 lactations. J. Dairy Sci. 95:918–924. https://doi.org/10.3168/jds.2011-4523.

Johnston, R., K. Jones, and D. Manley. 2018. Confounding and collinearity in regression analysis: A cautionary tale and an alternative procedure, illustrated by studies of British voting behaviour. Qual. Quant. 52:1957–1976. https://doi.org/10.1007/s11135-017-0584-6.

Köck, A., M. Ledinek, L. Gruber, F. Steininger, B. Fuerst-Waltl, and C. Egger-Danner. 2018. Genetic analysis of efficiency traits in Austrian dairy cattle and their relationships with body condition score and lameness. J. Dairy Sci. 101:445–455. https://doi.org/10.3168/jds.2017-13281.

Lembeye, F., N. López-Villalobos, J. L. Burke, S. R. Davis, J. Richardson, N. W. Sneddon, and D. J. Donaghy. 2016. Comparative performance in Holstein-Friesian, Jersey and crossbred cows milk once daily under a pasture-based system in New Zealand. N. Z. J. Agric. Res. 59:351–362. https://doi.org/10.1080/00288233.2016.1207669.

Ma, L., J. B. Cole, Y. Da, and P. M. VanRaden. 2019. Symposium review: Genetics, genome-wide association study, and genetic improvement of dairy fertility traits. J. Dairy Sci. 102:3735–3743. https://doi.org/10.3168/jds.2018-15269.

Macdonald, K. A., G. A. Verkerk, B. S. Thorrold, J. E. Pryce, J. W. Penno, L. R. McNaughton, L. J. Burton, J. A. S. Lancaster, J. H. Williamson, and C. W. Holmes. 2008. A comparison of three strains of Holstein-Friesian grazed on pasture and managed under different feed allowances. J. Dairy Sci. 91:1693–1707. https://doi.org/10.3168/jds.2007-0441.

Magne, M. A., and J. Quénon. 2021. Dairy crossbreeding challenges the French dairy cattle sociotechnical regime. Agron. Sustain. Dev. 41: 25. https://doi.org/10.1007/s13593-021-00683-2.

Malchiodi, F., A. Cecchinato, and G. Bittante. 2014a. Fertility traits of purebred Holsteins and 2- and 3-breed crossbred heifers and cows obtained from Swedish Red, Montbéliarde, and Brown Swiss sires. J. Dairy Sci. 97:7916–7926. https://doi.org/10.3168/jds.2014-8156.

Malchiodi, F., A. Cecchinato, M. Penasa, C. Cipolat-Gotet, and G. Bittante. 2014b. Milk quality, coagulation properties, and curd firmness modeling of purebred Holsteins and first- and second-generation crossbred cows from Swedish Red, Montéliarde, and Brown Swiss bulls. J. Dairy Sci. 97:4530–4541. https://doi.org/10.3168/jds.2013-7868.

Moraes, L. E., E. Kebreab, A. B. Strathe, J. Dijkstra, J. France, D. P. Casper, and J. G. Fadel. 2015. Multivariate and univariate analysis of energy balance data from lactating dairy cows. J. Dairy Sci. 98:4012–4029. https://doi.org/10.3168/jds.2014-8995.

NASEM (National Academies of Sciences, Engineering, and Medicine). 2021. Nutrient Requirements of Dairy Cattle. 8th rev. ed. National Academies Press.

NRC. 1996. Nutrient Requirements of Beef Cattle. 7th rev. ed. National Academies Press.

NRC. 2016. Nutrient Requirements of Beef Cattle. 8th rev. ed. National Academies Press.

O'Sullivan, M., P. Dillon, K. O'Sullivan, K. M. Pierce, N. Galvin, M. Egan, and F. Buckley. 2019. Intake, efficiency, and feeding behavior characteristics of Holstein-Friesian cows of divergent Economic Breeding Index evaluated under contrasting pasture-based feeding treatments. J. Dairy Sci. 102:8234–8246. https://doi.org/10.3168/jds.2019-16371.

Oldham, J. D., and G. C. Emmans. 1990. Animal performance as the criterion for feed evaluation. Pages 73–90 in Feedstuff Evaluation. J. Wiseman and D. J. A. Cole, ed. Butterworths.

Oltenacu, P. A., and D. M. Broom. 2010. The impact of genetic selection for increased milk yield on the welfare of dairy cows. Anim. Welf. 19(Suppl. 1):39–49. https://doi.org/10.1017/S0962728600002220.

Pereira, G. M., B. J. Heins, B. Visser, and L. B. Hansen. 2022. Comparison of 3-breed rotational crossbreds of Montbéliarde, Viking Red, and Holstein with Holstein cows fed 2 alternative diets for dry matter intake, production, and residual feed intake. J. Dairy Sci. 105:8989–9000. https://doi.org/10.3168/jds.2022-21783.

Piazza, M., M. Berton, N. Amalfitano, G. Bittante, and L. Gallo. 2023. Cull cow carcass traits and risk of culling of Holstein cows and 3-breed rotational crossbred cows from Viking Red, Montbéliarde, and Holstein bulls. J. Dairy Sci. 106:312–322. https://doi.org/10.3168/jds.2022-22328.

Piazza, M., D. Giannuzzi, R. Tessari, E. Fiore, M. Gianesella, S. Pegolo, S. Schiavon, E. Trevisi, F. Piccioli-Cappelli, A. Cecchinato, and L. Gallo. 2022. Associations between ultrasound hepatic measurements, body measures, and milk production traits in Holstein cows. J. Dairy Sci. 105:7111–7124. https://doi.org/10.3168/jds.2021-21582.

Prendiville, R., K. M. Pierce, and F. Buckley. 2009. An evaluation of production efficiencies among lactating Holstein-Friesian, Jersey, and Jersey × Holstein-Friesian cows at pasture. J. Dairy Sci. 92:6176–6185. https://doi.org/10.3168/jds.2009-2292.

Roche, J. R., N. C. Friggens, J. K. Kay, M. W. Fisher, K. J. Stafford, and D. P. Berry. 2009. Invited review: Body condition score and its association with dairy cow productivity, health, and welfare. J. Dairy Sci. 92:5769–5801. https://doi.org/10.3168/jds.2009-2431.

Saha, S., N. Amalfitano, G. Bittante, and L. Gallo. 2020. Milk coagulation traits and cheese yields of purebred Holsteins and 4 generations of 3-breed rotational crossbred cows from Viking Red, Montbéliarde, and Holstein bulls. J. Dairy Sci. 103:3349–3362. https://doi.org/10.3168/jds.2019-17576.

Schiavon, S., and G. Bittante. 2012. Double-muscled and conventional cattle have the same net energy requirements if these are related to mature and current body protein mass, and to gain composition. J. Anim. Sci. 90:3973–3987. https://doi.org/10.2527/jas.2011-4314.

Shonka-Martin, B. N., A. R. Hazel, B. J. Heins, and L. B. Hansen. 2019a. Three-breed rotational crossbreds of Montbéliarde, Viking Red, and Holstein compared with Holstein cows for dry matter intake, body traits, and production. J. Dairy Sci. 102:871–882. https://doi.org/10.3168/jds.2018-15318.

Shonka-Martin, B. N., B. J. Heins, and L. B. Hansen. 2019b. Three-breed rotational crossbreds of Montbéliarde, Viking Red, and Holstein compared with Holstein cows for feed efficiency, income over feed cost, and residual feed intake. J. Dairy Sci. 102:3661–3673. https://doi.org/10.3168/jds.2018-15682.

Sørensen, M. K., E. Norberg, J. Pedersen, and L. G. Christensen. 2008. Invited review: Crossbreeding in dairy cattle: A Danish perspective. J. Dairy Sci. 91:4116–4128. https://doi.org/10.3168/jds.2008-1273.

VandeHaar, M. J., and N. St-Pierre. 2006. Major advances in nutrition: Relevance to the sustainability of the dairy industry. J. Dairy Sci. 89:1280–1291. https://doi.org/10.3168/jds.S0022-0302(06)72196-8.

Viking Genetics. 2023. VikingRed: Healthy, trouble-free cows. Accessed Jan. 4, 2023. https://innovativebreeding.vikinggenetics.com/brochures/breed-brochures/vikingred.

Waltner, S. S., J. P. McNamara, J. K. Hillers, and D. L. Brown. 1994. Validation of indirect measures of body fat in lactating cows. J. Dairy Sci. 77:2570–2579. https://doi.org/10.3168/jds.S0022-0302(94)77198-8.

Yang, C. T., C. M. Wang, Y. G. Zhao, T. B. Chen, A. Aubry, A. W. Gordon, and T. Yan. 2020. Updating maintenance energy requirements for the current sheep flocks and the associated effect of nutritional and animal factors. Animal 14:295–302. https://doi.org/10.1017/S1751731119002064.

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