Metabolic parameters in transition cows as indicators for early-lactation culling risk

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ABSTRACT

A retrospective cohort study was performed with the objective of determining whether the serum concentrations of nonesterified fatty acids (NEFA), β -hydroxybutyric acid (BHBA), or calcium were associated with the risk of culling within 60 d in milk (DIM) in Holstein cows, and to establish thresholds for each metabolite that were predictive of increased culling risk. Data from 5,979 cows in Ontario (Canada) and several US states were obtained from 4 previously reported studies. For each metabolite and each of 3 sampling weeks (−1, +1, and +2 relative to calving), an optimal threshold was calculated based on having the maximum combined sensitivity (Se) and specificity (Sp) and used to categorize the serum concentrations into high and low risk groups. Logistic regression models were built for each metabolite and each week of sampling, as well as considering together all metabolites in wk −1 and wk +1 relative to calving. Cow was considered the experimental unit and herd as a random effect. Considered separately, precalving NEFA ≥ 0.4 mmol/L [odds ratio (OR) = 1.8; 95% confidence interval (CI) = 1.4 to 2.2], NEFA ≥ 0.8 mmol/L in wk +1 relative to calving (OR = 2.0; 95% CI = 1.5 to 2.6) and NEFA ≥ 0.8 mmol/L in wk +2 (OR = 4.2; 95% CI = 1.9 to 9.4 for cows in lactation 2; OR = 2.1; 95% CI = 1.4 to 3.3 for cows in lactation ≥ 3) were each associated with an increased risk of culling within the first 60 DIM. Similarly, BHBA ≥ 0.7 mmol/L in wk −1 (OR = 1.8; 95% CI = 1.3 to 2.5), BHBA ≥ 1.2 mmol/L in wk +1 (OR = 1.8; 95% CI = 1.4 to 2.2), and BHBA ≥ 1.6 mmol/L in wk +2 (OR = 3.2; 95% CI = 1.6 to 6.4 for cows in lactation 2; OR = 2.3; 95% CI = 1.6 to 3.3 for cows in lactation ≥ 3) were each associated with an increased risk of culling within the first 60 DIM. Likewise, calcium ≤ 2.3 mmol/L in wk −1 (OR = 1.6; 95% CI = 1.2 to 2.2), calcium ≤ 2.2 mmol/L in wk +1 (OR

= 1.5; 95% CI = 1.2 to 1.9), and calcium ≤ 2.3 mmol/L in wk +2 (OR = 2.3; 95% CI = 1.1 to 3.1) were each associated with an increased risk of culling within the first 60 DIM. When all metabolites were analyzed together, serum NEFA and calcium concentrations in wk −1 and serum NEFA concentration in wk +1 remained in the models. In conclusion, elevated serum NEFA and BHBA concentrations and lower serum calcium concentrations within 1 wk before calving through 2 wk after calving were associated with an increased risk of culling in early lactation. Measuring the concentration of selected metabolites around parturition may help to develop monitoring and intervention strategies to prevent early culling in transition dairy cows.

Key words: nonesterified fatty acids, β -hydroxybutyric acid, hypocalcemia, culling

INTRODUCTION

The transition period of a dairy cow, defined as 3 wk before calving until 3 wk after calving, is critical in that it is associated with a peak incidence of disease, including those of metabolic, nutritional, or infectious nature (Mulligan and Doherty, 2008). Up to 50% of dairy cows in the transition period may be affected by an infectious or metabolic disease (LeBlanc, 2010). Diseases associated with changes taking place during the transition period include, but are not limited to, retained placenta (Cameron et al., 1998), metritis (Hammon et al., 2006), endometritis (Dubuc et al., 2010), displaced abomasum (LeBlanc et al., 2005), ketosis (Green et al., 1999), and milk fever (Horst et al., 1994). These diseases have been generally associated with increased culling at a time or price that may not be economically optimal and often represent a financial loss (Gröhn et al., 1998; Fetrow et al., 2006). Hadley et al. (2006) analyzed a large data set from various regions of the United States and found that the average culling rate, excluding cows sold to another dairy, was 31.6%. Of these culling events, 79.5% were because of health issues. Dairy cows are predisposed to disease in the transition period as a result of a variety of factors, including reduced feed

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intake and high energy output resulting in a state of negative energy balance, lipolysis, and weight loss in early lactation; hypocalcemia in the few days postcalving; immune suppression for 1 to 2 wk before and 2 to 3 wk after calving; and bacterial contamination of the uterus for 2 to 3 wk after calving (Goff, 2008; LeBlanc, 2010). In addition, these cows also experience dietary changes and environmental stressors such as regrouping (Mulligan and Doherty, 2008).

Circulating concentrations of NEFA and BHBA measure aspects of the success in adaptation to a state of negative energy balance (Herdt, 2000; LeBlanc 2010). The concentration of serum NEFA reflects the magnitude of fat mobilization, whereas the concentration of BHBA reflects the completeness of oxidation of fat in the liver (LeBlanc, 2010). Serum calcium concentrations reflect the ability of a cow to replace extracellular calcium lost to colostrum and milk production by withdrawing calcium from bone and by increasing the efficiency with which dietary calcium is absorbed (Horst et al., 1994).

Several studies have determined NEFA and BHBA thresholds for predicting disease in transition dairy cattle (Cameron et al., 1998; LeBlanc et al., 2005; Ospina et al., 2010a). Fewer studies have examined metabolite concentrations as they relate to culling risk. Seifi et al. (2011) defined the thresholds for culling risk for calcium and NEFA in the first week postcalving as ≤ 2.2 mmol/L and ≥ 1.0 mmol/L, and in the second week postcalving as ≤ 2.3 mmol/L and ≥ 1.0 mmol/L, respectively.

The objectives of this retrospective cohort study were (1) to determine whether serum concentrations

of NEFA, BHBA, and calcium around parturition are associated with an increased risk of early-lactation culling (within 60 DIM), and (2) to determine optimal thresholds for each metabolite that are predictive of the risk of culling in early lactation.

MATERIALS AND METHODS

Data Collection

Data from 5,979 dairy cows located in various regions of Canada and the United States were obtained from 4 previously reported clinical trials (Table 1). Data sets contained information about the serum concentrations of NEFA, BHBA, and calcium for 3 sampling periods: wk -1 , wk $+1$, and wk $+2$ relative to calving. In all studies, cows were followed for a minimum of 60 d following parturition to collect additional information pertaining to disease and culling occurrence. Hence, despite some of the studies collecting events beyond this time, only data events recorded within 60 DIM were considered accurate and consistent between studies and were therefore used for this study. Diseases consistently recorded across all studies included milk fever, retained placenta, displaced abomasum, clinical ketosis, and metritis.

Data Analysis

Data were imported into SAS version 9.2 (SAS Institute Inc., Cary, NC) for statistical analysis. The cow was considered the experimental unit for all analyses. For each metabolite and week of sampling (wk -1 , wk

Table 1. Data sources used in pooled analysis of metabolic risk factors for culling in early lactation

Variable	Duffield et al. (1998)	LeBlanc et al. (2005)	Dubuc et al. (2010)	Chapinal et al. (2011)
Cows (no.)	996	1,112	1,649	2,222
Herds (no.)	25	20	3	21
Culling within 60 DIM (%)	10.0	7.4	6.7	8.8
Regions in which the study was conducted ¹	ON	ON	NY, ON	CA, FL, GA, MN, NC, NY, ON, SC, VA, WI
Sample time relative to calving/metabolite				
Wk -1				
NEFA		X	X	X
BHBA		X	X	X
Calcium	X	X		X
Wk $+1$				
NEFA		X	X	X
BHBA	X	X	X	X
Calcium	X	X		X
Wk $+2$				
NEFA			X	X
BHBA	X		X	X
Calcium	X			X

¹CA = California; FL = Florida; GA = Georgia; MN = Minnesota; NC = North Carolina; NY = New York; ON = Ontario, Canada; SC = South Carolina; VA = Virginia; WI = Wisconsin.

+1, and wk +2 relative to calving), a series of incremental thresholds of concentrations was created to be tested for association with culling risk within 60 DIM. Initial screening of thresholds for simple association with culling was performed using 2×2 contingency tables and chi-squared tests (PROC FREQ). Among associations with $P < 0.05$, the maximum combined sensitivity (**Se**) and specificity (**Sp**) was chosen to be the optimal threshold and used to categorize the concentrations of NEFA, BHBA, and calcium into high and low risk groups. Univariable associations between culling and all cow-level covariates (lactation category; season of calving; BCS; occurrence of twins, dystocia, retained placenta, ketosis, displaced abomasum, milk fever, metritis; and experimental treatment where appropriate) were also tested using chi-squared tests. Lactation was categorized into 1, 2, or ≥ 3 ; calving season into warm (May through August) and cool (September through April); and BCS into thin (≤ 3.0), moderate (3.25 to 3.75), and fat (≥ 4.0). Variables with $P > 0.20$ in the initial screening were not considered further.

Separate multivariable logistic regression models (PROC GLIMMIX with binary distribution and logit link function) were first created for each metabolite and week of sampling, including the dichotomized metabolite, the covariates with $P \leq 0.20$ in the univariable analysis, and the biologically plausible 2-way interactions. Herd within study and study were considered as random effects. Ketosis and displaced abomasum were considered intervening variables for models containing NEFA and BHBA, and milk fever for models containing calcium, and therefore were not included in those models. Precalving models for each metabolite were re-run to exclude all variables that would not be known at the time of wk -1 sample collection (occurrence of twins, dystocia, and clinical disease), as an approach with practical application. Covariates and interactions were removed from the final model if $P > 0.05$ by manual backward selection. To avoid confounding, covariates remained in the final model if their removal caused $\geq 20\%$ change in the estimate of the main predictors on the log odds scale. Residuals were examined for each model to detect possible outliers and influential observations.

For wk -1 and wk +1, a similar multivariable logistic model was built including the 3 dichotomized metabolites together. A multivariable logistic model including all metabolites in wk +2 could not be created because the number of studies that collected data on all metabolites in wk +2 was too small. Spearman correlation coefficients between the 3 dichotomized metabolites within the same week were calculated to avoid submitting highly correlated variables to the same model. The correlation coefficients were ≤ 0.36 in all cases.

RESULTS

In the univariable analyses, we found associations between culling within 60 DIM and lactation category [odds ratio (**OR**) = 0.6; 95% CI = 0.5–0.7; $P = 0.12$, when comparing lactation 1 against ≥ 3 ; OR = 0.5; 95% CI = 0.4–0.6; $P < 0.0001$, when comparing lactation 2 against ≥ 3]; occurrence of twins (OR = 1.4; 95% CI = 1.1–1.8; $P = 0.01$), dystocia (OR = 1.5; 95% CI = 1.1–1.9; $P = 0.01$), retained placenta (OR = 1.5; 95% CI = 1.2–2.0; $P = 0.002$), clinical ketosis (OR = 2.5; 95% CI = 1.9–3.4; $P < 0.0001$), displaced abomasum (OR = 3.6; 95% CI = 2.6–4.9; $P < 0.0001$), milk fever (OR = 2.2; 95% CI = 1.5–3.3; $P < 0.0001$); and season of calving (OR = 1.2; 95% CI = 1.0–1.5; $P = 0.03$, when comparing warm against cool calving season).

NEFA

The optimal NEFA threshold in wk -1 relative to calving for prediction of culling risk within 60 DIM as defined by the maximum sum of Se and Sp was ≥ 0.4 mmol/L (Se = 53%, Sp = 63%). Controlling for the effects of lactation category and dystocia, NEFA ≥ 0.4 mmol/L was associated with culling risk (Table 2). When the model was re-run to exclude dystocia, coefficients for the main predictor and covariate changed by less than 5%.

The optimal NEFA threshold for prediction of culling risk was ≥ 0.8 mmol/L in wk +1 and wk +2 relative to calving (Se = 53%, Sp = 61%, and Se = 49%, Sp = 61%, respectively). The model for NEFA in wk +1, controlling for lactation category and milk fever, showed an association between serum NEFA ≥ 0.8 mmol/L and culling risk. The model for serum NEFA in wk +2, controlling for milk fever, revealed a NEFA by lactation category interaction ($P = 0.02$). When the model was stratified by lactation category, NEFA was only associated with risk of culling in cows in second lactation and greater.

BHBA

The optimal BHBA threshold in wk -1 relative to calving for prediction of early-lactation culling risk was ≥ 0.7 mmol/L (Se = 29%, Sp = 82%). In a logistic model controlling for lactation category and dystocia, serum BHBA ≥ 0.7 mmol/L was associated with an increased risk of culling (Table 2). When the model was re-run to exclude dystocia, coefficients for the main predictor and covariate changed by less than 5%.

The optimal BHBA threshold in wk +1 relative to calving for predicting culling risk within 60 DIM was ≥ 1.2 mmol/L (Se = 31%, Sp = 82%). Controlling for

Table 2. Logistic regression models of the association of serum metabolites measured once in each of wk -1, wk +1, and wk +2 relative to calving in Holstein cows with the risk of culling within 60 DIM, when each metabolite was tested in a separate model for each week¹

Variable	Samples (no.)	Animals in the high risk group (%)	Odds ratio	95% CI	P-value
Wk -1					
NEFA ≥ 0.4 mmol/L	4,742	38.3	1.8	1.4-2.2	<0.0001
BHBA ≥ 0.7 mmol/L	3,152	18.9	1.8	1.3-2.5	<0.0001
Calcium ≤ 2.3 mmol/L	3,086	22.1	1.6	1.2-2.2	0.0009
Wk +1					
NEFA ≥ 0.8 mmol/L	3,249	40.5	2.0	1.5-2.6	<0.0001
BHBA ≥ 1.2 mmol/L	5,793	19.3	1.8	1.4-2.2	<0.0001
Calcium ≤ 2.2 mmol/L	4,134	45.0	1.5	1.2-1.9	0.002
Wk +2					
NEFA ≥ 0.8 mmol/L ²	2,148	22.1			
Lactation 1			0.6	0.2-1.7	0.32
Lactation 2			4.2	1.9-9.4	0.001
Lactation ≥ 3			2.1	1.4-3.3	0.001
BHBA ≥ 1.6 mmol/L ³	5,793	11.8			
Lactation 1			0.7	0.3-1.8	0.46
Lactation 2			3.2	1.6-6.4	0.001
Lactation ≥ 3			2.3	1.6-3.3	<0.0001
Calcium ≤ 2.3 mmol/L	3,032	43.0	2.3	1.1-3.1	<0.0001

¹Metabolite concentrations were dichotomized at different thresholds and only the threshold with the maximum combined sensitivity and specificity are shown. Models accounted for the effect of lactation category, dystocia, and clinical diseases when these variables were significant.

²Lactation by NEFA interaction ($P = 0.02$).

³Lactation by BHBA interaction ($P = 0.03$).

lactation category and dystocia, BHBA ≥ 1.2 mmol/L was associated with an increased culling risk.

The optimal BHBA threshold in wk +2 for prediction of culling risk was ≥ 1.6 mmol/L (Se = 21%, Sp = 89%). The logistic model for BHBA in wk +2 revealed an interaction between BHBA and lactation category ($P = 0.03$). When the model was stratified by lactation category, BHBA was only associated with risk of culling in cows in second lactation or greater.

Calcium

The optimal calcium threshold in wk -1 relative to calving for predicting culling risk within 60 DIM was ≤ 2.3 mmol/L (Se = 33%, Sp = 78%). The precalving model showed that serum calcium ≤ 2.3 mmol/L was significantly associated with culling risk, controlling for lactation category, dystocia, ketosis, and displaced abomasum (Table 2). When the model was re-run to exclude displaced abomasum, ketosis, and dystocia, coefficients for the main predictor and covariate changed by less than 5%.

The optimal calcium threshold in wk +1 relative to calving for predicting culling risk was ≤ 2.2 mmol/L (Se = 57%, Sp = 56%). When controlling for lactation category, clinical ketosis, and displaced abomasum, the model for wk +1 showed that serum calcium ≤ 2.2 mmol/L was associated with increased culling risk.

The optimal calcium threshold in wk +2 relative to calving for predicting culling risk was ≤ 2.3 mmol/L (Se = 63%, Sp = 59%). The model for wk +2 showed that serum calcium ≤ 2.3 mmol/L was associated with culling risk, controlling for lactation category, ketosis, and displaced abomasum.

All Metabolites

When NEFA, BHBA, and calcium concentrations in wk -1 were submitted to the same multivariable logistic model, NEFA ≥ 0.4 mmol/L and calcium ≤ 2.3 mmol/L remained in the model, after accounting for the effects of lactation category and dystocia (Table 3). When NEFA, BHBA, and calcium concentrations in wk +1 were submitted to the same multivariable logistic model, only NEFA ≥ 0.8 mmol/L remained in the model, after accounting for the effect of lactation category (Table 4).

DISCUSSION

The results of this study highlight the association of the metabolic parameters NEFA, BHBA, and calcium with culling risk in early lactation. These results are in agreement with a report by Seifi et al. (2011) on the predictive association of elevated NEFA and BHBA and reduced calcium in the transition period

Table 3. Logistic regression model of the association of the serum metabolites measured in wk -1 relative to calving in Holstein cows ($n = 3,086$) with the risk of culling before 60 DIM, when considered in the same model

Variable	Estimate	SE	<i>P</i> -value	Odds ratio	95% CI
Lactation					
1	-0.82	0.17	<0.0001	0.4	0.3–0.6
2	-0.84	0.18	<0.0001	0.4	0.3–0.6
≥ 3	Referent				
Dystocia	0.53	0.21	0.01	1.7	1.1–2.6
NEFA ≥ 0.4 mmol/L	0.64	0.14	<0.0001	1.9	1.4–2.5
Calcium ≤ 2.3 mmol/L	0.53	0.21	0.01	1.7	1.1–2.6

with subsequent culling in early lactation. Lower DMI in conjunction with increased energy and nutrient demands required for lactation results in increased lipid mobilization and increased production of ketones and, in addition to increased the calcium output required for colostrum and high milk production, are characteristic of the transition period (Goff, 2008; LeBlanc, 2010). These factors translate to elevated NEFA and BHBA and lower calcium levels, which have previously been shown to be associated with increased risk of clinical disease (LeBlanc et al., 2005; Seifi et al., 2011), lower milk yield (Ospina et al., 2010b), and impaired reproductive performance (Walsh et al., 2007). Culling is affected by the cost of replacing the animal, as well as a variety of other factors including disease, reproduction problems, and milk production (Gröhn et al., 1998; Fetrow et al., 2006; Hadley et al., 2006). These factors may stem from elevated serum NEFA and BHBA and lower calcium concentrations (Ospina et al., 2010a,b). Gröhn et al. (1998) found that higher milk production represented a protective effect against culling, and that milk fever, retained placenta, displaced abomasum, and clinical ketosis all significantly affected culling at different stages of lactation.

The thresholds determined in this study for predicting early lactation culling risk are similar to values suggested in other studies for predicting disease and culling risk in transition dairy cows. Similar to our results, Hammon et al. (2006) reported that NEFA ≥ 0.4 mmol/L before parturition (wk -2 and -1) was associated with reduced peripheral blood neutrophil function and higher risk of metritis or subclinical endometritis.

Other studies reported precalving NEFA thresholds between 0.3 and 0.5 mmol/L for association with metritis, retained placenta, and displaced abomasum (Cameron et al., 1998; LeBlanc et al., 2005; Chapinal et al., 2011). Similar to the present threshold of NEFA ≥ 0.8 mmol/L in wk $+1$, LeBlanc et al. (2005) and Chapinal et al. (2011) reported a threshold of ≥ 1.0 mmol/L for predicting displaced abomasum. Seifi et al. (2011) also reported a postcalving threshold for NEFA of ≥ 1.0 mmol/L as a predictor of culling in the first 2 mo of lactation. Ospina et al. (2010a) reported a lower postcalving NEFA threshold of 0.57 mmol/L for predicting any of displaced abomasum, clinical ketosis, metritis, or retained placenta. Differences in results between our study and Ospina et al. (2010a) may be a reflection of differing sampling times. Ospina et al. (2010a) obtained the precalving sample within 2 wk before calving and the postcalving sample within 2 wk after calving. The critical threshold for BHBA in wk -1 of ≥ 0.7 mmol/L determined in this study for predicting culling risk was close to the threshold of ≥ 0.8 mmol/L reported by Chapinal et al. (2011) for predicting displaced abomasum. Duffield et al. (2009), LeBlanc et al. (2005), and Seifi et al. (2011) all reported the same threshold as determined in this study of ≥ 1.2 mmol/L BHBA in wk $+1$ for the prediction of various diseases. Ospina et al. (2010a) reported a slightly lower threshold for BHBA in wk $+1$ postcalving of ≥ 1.0 mmol/L for predicting any of displaced abomasum, clinical ketosis, metritis, or retained placenta, which might be a reflection of the wider sampling timeframe before and following calving, as well as the broader outcome of any of 3

Table 4. Logistic regression model of the association of the serum metabolites measured in wk $+1$ relative to calving in Holstein cows ($n = 3,249$) with the risk of culling before 60 DIM, when considered in the same model

Variable	Estimate	SE	<i>P</i> -value	Odds ratio	95% CI
Lactation					
1	-0.70	0.17	<0.0001	0.5	0.4–0.7
2	-0.79	0.18	<0.0001	0.5	0.3–0.6
≥ 3	Referent				
NEFA ≥ 0.8 mmol/L	0.68	0.14	<0.0001	2.0	1.5–2.6

diseases. Duffield et al. (2009) reported a threshold for BHBA in wk +2 of ≥ 1.8 mmol/L for predicting an increased risk of displaced abomasum, which is similar to the threshold here of ≥ 1.6 mmol/L in wk +2 for predicting culling risk. Chapinal et al. (2011) also determined the same precalving calcium threshold of ≤ 2.2 mmol/L as in this study for predicting displaced abomasum. The calcium threshold of ≤ 2.2 mmol/L in wk +1 after calving for predicting culling risk is similar to that reported by Seifi et al. (2011) of ≤ 2.3 mmol/L for predicting displaced abomasum. However, a threshold of ≤ 2.2 mmol/L around parturition is higher than expected for undesirable outcomes, as it is within the range (1.64–2.61 mmol/L) reported for cows sampled in the first week after calving that had no clinical disease in the transition period (Quiroz-Rocha et al., 2009). Suboptimal calcium concentrations may be a reflection of inadequate DMI.

The magnitude of the measure of association (OR = 2.0, 95% CI = 1.5–2.6) of NEFA concentration in the first week after calving with culling risk was smaller than that reported by Seifi et al. (2011), in which the odds of culling were nearly 5 times greater if serum NEFA in the week after calving was ≥ 1.0 mmol/L (95% CI = 2.3–9.6). Despite the numerical difference in the estimate, the CI reported by Seifi et al. (2011) for this association were much wider and overlapped with those reported in our study. Only NEFA remained a significant predictor for culling risk in early lactation when serum NEFA, BHBA, and calcium in wk +1 relative to calving were all submitted to the same model. This is similar to Ospina et al. (2010a), where NEFA remained the only predictor of disease when both postcalving NEFA and BHBA in wk +1 and +2 were submitted to the same multivariable logistic regression model. This is interesting because previous research has typically focused on precalving NEFA and postcalving BHBA concentrations. Associations of postcalving serum NEFA concentrations with disease and culling have not been as thoroughly investigated, and the results of this research warrant further investigation on the usefulness of postcalving NEFA as a predictor of disease and culling. Postcalving NEFA has also been shown to have a stronger association with milk production than BHBA. When Ospina et al. (2010b) submitted both postcalving NEFA and BHBA (wk +1 and +2) to the same model for predicting milk production, only NEFA remained in the model.

Serum BHBA was a significant predictor of culling risk when submitted as the only metabolic variable in a logistic regression model for wk –1 relative to calving. This is interesting, because NEFA has been the primary metabolic parameter of interest before calving. The findings of our study suggest the possible utility

of BHBA in the week before calving as a metabolic predictor of disease and culling risk, particularly due to the availability of a practical, validated cow-side test for measuring blood BHBA (Iwersen et al., 2009). On the other hand, in the multivariable analysis for wk –1, BHBA did not remain in the model when NEFA and calcium were included.

Although periparturient concentrations of NEFA seem to be more strongly associated with culling than BHBA, BHBA was associated with culling when assessed separately. The 2 metabolites were not highly correlated, which means that cows at risk of culling might not show high levels of both NEFA and BHBA at the same time. Given the logistic difficulties of monitoring both parameters frequently in the transition period, and given the fact that cow-side tests to measure BHBA are available, measuring both parameters once in the week before and in the week after calving would likely increase the probability of detecting cows at risk of early lactation culling.

The magnitude of the measures of association of decreased calcium concentration in the first and second week postcalving with culling risk (OR = 1.5 and 2.3, respectively) were smaller than those reported by Seifi et al. (2011), in which the odds of culling were 3.2 times greater if calcium was ≤ 2.2 mmol/L in wk +1 relative to calving, and 6.5 times greater if calcium was ≤ 2.2 mmol/L in wk +2. The lower apparent magnitude of effect in the present analysis may be due to the wider variety of herds, regions, and management and nutritional practices in our data, which brings more unmeasured sources of variability to the analysis.

In addition to serum NEFA, BHBA, and calcium concentrations, several other covariates were examined in each model, including lactation category, calving season, and occurrence of twins, dystocia, and clinical disease. Lactation category was the only covariate that remained significant in all models. In agreement with Gröhn et al. (1998), older cows showed a higher risk of being culled than cows in lactation 1 and 2. In both logistic regression models for NEFA and BHBA in wk +2, we found an interaction between lactation and the metabolite concentration. Multiparous cows with high NEFA and BHBA had higher odds of being culled. In the case of primiparous cows, the odds of being culled were not affected by NEFA and BHBA concentrations in wk +2. Similarly, Ospina et al. (2010b) found an interaction between postpartum NEFA and BHBA concentrations and parity in the prediction of milk production. Whereas a high concentration of NEFA or BHBA in the 2 wk after calving was associated with milk loss in multiparous cows, it was associated with milk increase in primiparous cows. These results suggest that primiparous cows may have a different physiological

response to postcalving metabolic challenges, because they have to balance demands for growth with those for milk production and may mobilize fat more readily before its health and productivity are compromised.

CONCLUSIONS

Elevated serum NEFA or BHBA concentrations and lower serum calcium concentrations from 1 wk before calving to 2 wk after calving were associated with an increased risk of being culled within the first 60 DIM. Serum NEFA and calcium concentrations in the week before calving in combination were associated with an increased risk of culling. However, these indicators provided poor to fair sensitivity (many cows that were culled were not detected by these variables) and fair to good specificity (typically, few cows that were above the thresholds that assessed NEFA and BHBA or below those for calcium were not culled). This underlines that the risk of removal from the herd in early lactation is multifactorial and that these indicators were measured days to weeks before culling occurred. Measuring the concentration of selected metabolites around parturition may help to develop monitoring and intervention strategies to prevent the uneconomical culling of transition dairy cows.

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