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NON-NUTRITIONAL COW-LEVEL AND AMBIENT TEMPERATURE RISK FACTORS FOR THE OCCURRENCE OF LEFT DISPLACED ABOMASUM IN HOLSTEIN COWS

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ABSTRACT

This retrospective cohort study (n= 13411 lactations) aimed to evaluate the association between temperature-humidity index at calving and cow-related variables with the occurrence of left displaced abomasum (LDA) in Holstein cows in a hot environment. Variables eligible for multiple logistic models were age at first calving, lactation number, temperature-humidity index at calving, metritis, lactation commenced following an abortion, dystocic parturition, premature calving, twin births, body weight at parturition, body condition score (BCS) at parturition, BCS change between calving and first artificial insemination (AI), and wither height at parturition. Significant risk factors for LDA were number of calvings [5.2% for primiparous vs. 8.3% for multiparous cows; odds ratio (OR)=1.3, P < 0.01], BCS at calving (6.6% for BCS \geq 3.5 and 8.4 for BCS <3.5; OR=0.75, P < 0.01), BCS change from calving to first AI (7.2% for <0.85 and 9.2 for \geq 0.85 units; OR=1.5, P < 0.01) and lactation initiated with abortion (1.6 for abortion and 10.2 for no abortion; OR=0.2, P < 0.01). Heat stress at calving was not associated with LDA. It was concluded that BCS <3.5 at calving and a greater BCS loss from calving to the first AI were associated with an increased risk of LDA. In contrast, initiation of lactation with abortion was related to a lower incidence of LDA.

Keywords: abortion, body condition score, dystocic parturition, heat stress, metritis, left displaced abomasum.

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INTRODUCTION

Left displacement of the abomasum (LDA) is a metabolic disease in high-yielding dairy cattle during early lactation (Caixeta et al., 2018; Fiorentin et al., 2018). Therefore, direct economic losses (surgery, discarded milk, medication, or on-farm death) and indirect losses (subsequent milk yield reduction, body weight loss, decreased fertility, and shortening of the productive life of affected cows) represent substantial economic losses for dairy farmers (Caixeta et al., 2018).

The mean incidence of this metabolic disorder in dairy herds in the United States is approximately 3.5% (Constable et al., 2017). Although the mechanisms involved in abomasum displacement have not been completely elucidated, this disorder is multifactorial, with nutrition, stress conditions, and metabolic disturbances contributing to this disease (Fiore et al., 2019; Yong et al., 2021). Specific non-genetic factors include the occurrence of metabolic diseases, such as hypocalcemia and ketosis, which affect abomasal motility and interfere with abomasal emptying, which increases gas accumulation (Rodríguez et al., 2017; Neves et al., 2018; Kang et al., 2019) and reproductive disorders derived from parturition (Tschoner et al., 2022). Additional factors predisposing cows to develop LDA include advanced parity (Kang et al., 2019; Pérez-Báez et al., 2021), breed (Zerbin et al., 2015; Dyck et al., 2023), high pre-partum body condition score (BCS; Song et al., 2020), and pre-partum negative energy balance (Esposito et al., 2014; Pérez-Báez et al., 2021), and health conditions (mastitis, endometritis; Tschoner et al., 2022). Other influencing factors include nutritional practices such as high concentrate and low roughage diets (Behluli et al., 2017) and corn silage with reduced particle size (Simoes et al., 2013).

Heat stress has not been studied as a risk factor for LDA. However, this condition could affect this metabolic disease because heatstressed, high-producing cows have increased blood insulin levels, which results in decreased adipose tissue mobilization and increased glucose utilization by peripheral tissues (Mann, 2022). Additionally, heat stress reduces feed intake, decreases activity, and increases drinking (Garner et al., 2017), which increases plasma non-esterified fatty acids (NEFA) concentrations to maximize milk synthesis; thus, the energy homeostasis of heat-stressed cows is altered (Mann, 2022). Increasing the energy density of the diet is beneficial from the point of view of managing heat stress but puts cows at greater risk of displaced abomasum.

Although previous studies have provided ample information on the risk factors associated with displaced abomasum, significant gaps still existin our understanding of its pathogenesis.

Therefore, we hypothesized that thermal stress at the beginning of lactation and severe loss of BCS from calving to first breeding would increase the risk of displaced abomasum in Holstein cows in a hot environment. Therefore, the objectives of this study were to assess (1) the effect of heat stress at calving on the occurrence of LDA in transition Holstein cows, (2) the effect of BCS at calving and the change in BCS from calving to first artificial insemination (AI) on the occurrence of LDA, (3) the effects of calving-related events on LDA, and (4) the effect of other cow-specific risk factors for LDA.

MATERIAL AND METHODS

Animals, feeding, and facilities

Cows included in the present study were handled following the guidelines outlined by the 'Guide for Care and Use of Agricultural Animals in Research and Teaching' (3rd ed. 2010 Federation of Animal Science Societies, Champagne, IL; https://www.fass.org/images/science-policy/Ag_Guide_3rd_ed.pdf). The study was also approved by the Autonomous Agrarian University Antonio Narro Animal Care Advisory Board (# 3001-2419).

The study was conducted at a large commercial dairy farm (~ 3000 lactating cows) in northern Mexico (25 °N) between January 2015 and December 2021. The climate in the study site is characterized by constantly high daytime temperatures, with an average annual temperature of 23.9 °C. Maximum and minimum ambient temperatures during the study period were 42.6 and 2.1 °C, respectively, with an average annual precipitation of 230 mm.

The cows were kept in open pens with dry, uncompacted manure surfaces and sufficient shade, and the feeding areas were shaded by metal roofing. Cows were fed a total mixed ration based on alfalfa hay, corn silage, and grain concentrate (corn grain, soybean meal, cottonseed meal, and a mineral premix) twice daily in approximately equal quantities at ~0800 and 1600 h. The forage: concentrate ratio was 50:50, which met the requirements of lactating Holstein cows weighing 650 kg and producing 38 kg of 3.5% fat-corrected milk (NRC, 2001). TMR was subsequently adjusted to the lactation stage. Cows were fed as often as desired with approximately 5% feed refusal for maximum feed intake and were milked three times daily at 01:00, 09:00, and 17:00 h. BCS (1-5 scale, 0.25



points increment) was recorded at calving and the first artificial insemination (after 50 days post-calving).

Study design, disease recording, and sample collection

retrospective observational study included 13,411 lactations. The dairy farm veterinarian identified metritis from day 4 until 20 after calving; cows were monitored for metritis twice on a weekly basis, using rectal palpation of the genital tract. Abnormal vaginal secretion is associated with watery, purulent, brown-colored, and fetid discharge, independent of the rectal temperature (Garzon et al., 2022). LDA was diagnosed based on reduced appetite and depression together with a distinct, highpitched tympanic resonance ("ping") produced by hitting the left side of the abdomen between the 9th and 12th ribs for a more precise diagnosis; re-examination was performed 24 hours later. The right displaced abomasum and right torsion of the abomasum were omitted.

Lactation commenced following an abortion (evident visual signs of abortion, such as eviction of a lifeless fetus or the presence of fetal membranes and vaginal discharges) and was defined as an abortion in the non-lactation period with days in gestation less than 260 days or heifer aborting in late gestation. Dystocia was defined as the farm veterinarian's assistance during calving (slight or considerable assistance). Premature birth was defined as calves born alive at 270 days of gestation. In addition, the age at first lactation and occurrence of twin births were recorded.

Climatic data were obtained from a meteorological station located 1.7 km away from the dairy farm. Daily temperatures and relative humidity were recorded throughout the study period. The air temperature was recorded using a mercury thermometer placed under a shade, 1.5 m above the ground. With this information, the daily temperature-humidity index (THI) was calculated using the following equation (daily temperature in degrees Celsius; RH refers to maximum relative humidity):

THI = $(0.8 \times \text{temperature}) + ((\% \text{RH}/100) \times$ (temperature - 14.4)) + 46.4.

THIs at calving were grouped into three classes: no heat stress (\leq 70), heat stress (70-85), and severe heat stress (≥ 85).

Statistical analysis

Variables eligible for logistic models were age at first calving (<24 months vs. >24 months), lactation number (primiparous vs. multiparous), metritis (yes vs. no), abortion before calving (yes vs. no), dystocic parturition (yes vs. no), premature calving (yes vs. no), twin births (yes vs. no) body weight at parturition (<554 vs. >554 kg), BCS at parturition ($<3.5 vs. \ge 3.5 \text{ values}$) BCS change between calving and first AI (<0.81 vs. >0.81 values), wither height at parturition (>132 vs. ≥132 cm) and THI at parturition (≤70, 70-85 and >85 values). To analyze the risk factors contributing to the occurrence of LDA (binary trait), these potential explanatory variables and one-way interactions were included in a multiple logistic regression model for screening significant variables affecting LDA using the PROC LOGISTIC procedure of SAS (SAS Inst. Inc., Cary, NC, USA), applying a backward stepwise logistic procedure to eliminate all nonsignificant variables. These were continuously removed from the model using the Wald statistic criterion if the significance level was greater than 0.05. This procedure estimated odds ratios and 95% confidence intervals (CIs). Therefore, the final model included only the main effects and calving year as covariate.

RESULTS

The proportion of cows presenting LDA from 2015 to 2021 was 7.4 (993 positive animals to LDA based on percussion auscultation/12,418 total animals; 95% CI= 6.9-7.9). The median time from parturition to the LDA was 16.0 days. The risk-adjusted risk factors for LDA are listed in Table 1. Multiparous cows had a 1.3 times higher risk of LDA than primiparous cows. Cows with BCS \geq 3.5 values had a reduced risk (P < 0.01) of LDA, similar to cows with BCS < 3.5 values. When the BCS change from calving to first artificial insemination was >0.85, cows had 1.5 times (P < 0.01) the risk of LDA than cows with a lesser BCS loss between parturition and first breeding.

The odds of LDA in cows with a wither height >132 cm were 1.3 times higher (P < 0.01) than those in cows with a wither height <132 cm. The initiation of lactation following an abortion substantially decreased the risk of LDA (P < 0.001) compared to cows whose lactation derived from a normal parturition. There was an interaction between BCS at calving and BCS change between calving and AI. The lactational incidence of LDA in cows with BCS <3.5 and a BCS change >0.85 was three times higher than that in cows with BCS > 3.5 and a BCS change between calving and the first AI > 0.85 values (Fig. 1). Severe heat stress at calving (THI >85) was not associated with the LDA.

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Variables	LDA (%)	Odds ratio (OR)	95% CI OR	р
Parity				< 0.0001
Multiparous	699/8438 (8.3)	1.3	1.2 - 1.6	
Primiparous	294/4973 (5.2)	Reference		
BCS at calving ¹				< 0.0001
≥3.5	469/7158 (6.6)	0.75	0.7 - 0.9	
<3.5	524/6253 (8.4)	Reference		
Change in BCS ²				0.0001
>0.85	122/1390 (9.2)	1.5	1.2 - 1.9	
< 0.85	871/12091 (7.2)	Reference		
Wither height				< 0.0001
>132 cm	603/7676 (7.9)	1.3	1.2 - 1.5	
<132 cm	390/5735 (6.8)	Reference		
Abortion ³				< 0.0001
Yes	72/4390 (1.6)	0.2	0.1 - 0.2	
No	921/9021 (10.2)	Reference		

Table 1. Final multivariate logistic regression model for factors associated with the incidence of left displaced abomasum (LDA) in high-yielding Holstein cows in a hot environment.

DISCUSSION

The mean prevalence of LDA in the current study was slightly higher than that reported in other studies (Song et al., 2020). Studies from North America, Brazil, and Canada have reported a much lower prevalence of displaced abomasum (Dubuc and Denis-Robichaud, 2017; Dyck et al., 2023). Higher parity was associated with an increased incidence of LDA, which is consistent with the results of previous studies (Kang et al., 2019).

For many cows in the present study, heat stress was severe during calving (THI >85). However, heat stress was not related to the LDA. This response was unexpected as heat stress reduces feed intake, leading to LDA (Esposito et al., 2014). In addition, the displaced abomasum coincides with environmental heat stress (Quanz et al., 2022). The absence of an association between heat stress and LDA in the present study could be due to the concentrate not exceeding 50% of the total dry matter intake, which may have prevented the development of displaced abomasum (Vermunt, 2022).

The higher risk of developing LDA in cows with higher parity could partly be explained by the fact that primiparous cows typically maintain good BCS in late pregnancy, as they have not previously experienced lactation. Compared to

multiparous cows, BCS can be better maintained in the late gestation and early lactation periods, although they do not recuperate BCS post-nadir as effectively as multiparous cows (Gärtner et al., 2019). Furthermore, first-parity cows mobilize significantly less body energy reserves than second- and third-parity cows (Piñeyrúa et al., 2018) and develop a less severe state of negative energy balance (Oikawa et al., 2019), making them less prone to LDA, as negative energy balance prepartum (based on plasma non-esterified fatty acids) is a significant risk factor for LDA (Song et al., 2020). Additionally, multiparous cows present higher odds of developing hypocalcemia and ketosis (McArt and Neves, 2020; Ha et al., 2023), both of which are associated with displaced abomasum (Rodríguez et al., 2017) due to their shared etiology.

In the present study, cows with BCS ≥3.5 at calving had lower odds of having LDA. These results are in line with previous observations (Dawod et al., 2014), where cows with an average BCS ≤2.0 in the first 100 DIM had twice the incidence of metabolic diseases (including displaced abomasum) than those with BCS >2.0. Likewise, it has been observed that cows with LDA have lower BCS than healthy cows (Song et al., 2020). Additionally, cows that lose body condition during the dry period have increased odds of developing displaced abomasum (Daros

¹BCS= Body condition score (1 to 5 scale scoring system, 0.25 points increment).

²Body condition score change from calving to first artificial insemination.

³Lactation initiated with an abortion.

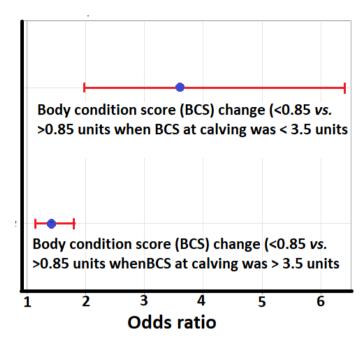


Fig. 1. Odds ratio for the occurrence of left displaced abomasum (LDA) in Holstein cows in a hot environment, as a function of body condition score change between calving and first artificial insemination, considering the body condition score at calving. Red lines indicate the 95% confidence intervals.

et al., 2020). These data suggest that cows with BCS ≥3.5 have an added active system for NEFA catabolism and glycerol utilization within the adipose tissue, as has been observed previously (Alharthi et al., 2018), in which reduced plasma NEFA in cows with BCS ≥3.5 at calving, would reduce LDA.

It has been postulated that cows with elevated BCS at parturition are more likely to develop metabolic diseases, including displacement of the abomasum (Caixeta et al., 2018; Rathbun et al., 2017) because over-conditioned cows tend to lose more body condition during early lactation than thin cows (Casaro et al., 2024). In the present study, only 2.6% of cows had a BCS of 4; therefore, the number of fat cows at calving was minimal, whereas most cows (83%) had a BCS between 2.5 and 3.5. Given that the optimum calving BCS for milk production and reproductive efficiency is 3.5 (Gobikrushanth et al., 2019), on a 5-point scale, cows with a BCS of 3.5 at parturition were not likely to experience severe fat mobilization and prolonged negative energy balance, which made less prone to have metabolic diseases and displaced abomasum.

Cows with higher BCS losses between calving and the first AI had greater odds of developing LDA than cows with lower BCS losses

postpartum. Changes in postpartum BCS are indirect indicators of fat mobilization and energy balance in individual cows (Alharthi et al., 2018) and are good predictors of the risk of postpartum metabolic diseases (Rodríguez et al., 2017). In the present study, cows that mobilized more tissue between calving and first AI likely experienced greater energy mobilization and a longer period of negative energy balance (NEB), which would increase the risk of LDA because cows unable to adapt to the challenging time at the beginning of lactation are more prone to have displaced abomasum (Coşkun et al., 2022). It is important to note that the effect of BCS loss postpartum on metabolic diseases can begin before parturition, as cows gaining BCS prepartum have a lesser reduction in BCS postpartum; therefore, BCS loss during the dry period can contribute to health disorders, including displaced abomasum (Chebel et al., 2018). In contrast, cows with minimal changes in BCS when entering the prepartum period present higher BCS loss postpartum (Wang et al., 2019) and higher blood β-hydroxybutyrate and NEFA (Barletta et al., 2017).

In the present study, lactations initiated with an abortion prevented LDA. This response is attributed to the lower milk yield in these cows (Keshavarzi et al., 2020), which suggests a lower mobilization of body reserves during the first month of lactation compared to cows with calving-derived lactation. Cows that had abortions while dry had a short dry period, and the absence of a dry period resulted in a lower peak milk yield. As a result, cows could quickly meet their energy requirements by feed intake (Kok et al., 2019). In cows without a dry period, metabolic status is improved, as indicated by increased energy balance, decreased plasma NEFA, and reduced liver triglyceride accumulation in the periparturient period (Kok et al., 2019).

In addition, feed intake was probably not drastically affected in cows with abortionderived lactation, as it happens in cows initiating a new lactation when lactating (no dry period) compared with cows with a standard dry period (Kok et al., 2019). Thus, aborted cows did not have a decreased energy intake or a drastic diet change in a short time, and presumably, cows did not have meaningful changes in the rumen microbiota, as they did not have to adjust to high-energy diets (Kumar et al., 2015). Therefore, in this scenario, cows with abortion-derived lactation probably had improved energy balance, decreased blood NEFA, and reduced liver triglycerides in the peri-abortion period in the subsequent lactation - conditions that did not lead to LDA. It is also possible that fetal loss prevented the uterus from sliding under the caudal rumen, thereby decreasing the reduction in rumen volume and preventing the displacement of the abomasum to the left side of the abdominal cavity. Additionally, dry matter intake probably did not decrease, which prevented a reduction in rumen fill, leading to more space for the abomasum to move and shift.

Stature was unfavorably associated with higher odds of LDA, suggesting that taller cows are more likely to develop LDA. This response is difficult to explain because digestive diseases, including LDA, are unrelated. Since primiparous cows were less likely to have LDA and these animals were shorter than mature cows, the stature effect is related to the reduced incidence of LDA in younger cows.

CONCLUSIONS

Data showed that higher parity increased the risk of left displaced abomasum (LDA). Therefore, proper feeding for peripartum pluriparous cows should be maintained to avoid severe metabolic disorders leading to LDA. In addition, attention should be paid to cows shortly before parturition to attain a BCS of 3.5 to reduce the risk of LDA; BCS loss ≥0.85 between calving and the first AI,

may constitute a warning of a postpartum risk of LDA in Holstein cows in this hot environment. Finally, as expected, new lactation derived from abortion prevented LDA, as these cows did not have excessive lipolysis and metabolic disturbances typically associated with calving-derived lactations.

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Author contributions

Conceptualization, Miguel Mellado, and José Eduardo García; Design, Jesús Mellado, and Lina Estrella Morgado; Supervision, Miguel Mellado; Resource, Leonel Avendaño Reyes, and Ulises Macías Cruz; Materials, Lina Estrella Morgado; Data collection and/or processing, Lina Estrella Morgado, and Jesús Mellado; Analysis and/or interpretation, Miguel Mellado; Literature Search, Ulises Macías Cruz; Writing, Miguel Mellado; Critical review, Jesús Mellado, and Ulises Macías Cruz.

Declaration of Interests

The authors declare that they have no financial or personal relationships that may have inappropriately influenced the writing of this article.

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