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Summary

The objectives of this retrospective cohort study were to determine the association of heat stress exposure during the transition period with production, health, reproduction, and survival during the first 90 days postpartum in dairy cows. A total of 5,722 Holstein cows (2,324 heifers and 3,397 cows) were categorized into environmental condition (EC) groups based on average temperature humidity index (THI) exposure as thermoneutral (TN) or heat stress (HS) during the pre (Pre) and early postpartum (Post) periods into TN-TN, TN-HS, HS-TN, and HS-HS. In heifers, exposure to HS during the Pre, Post, or Pre and Post was associated with a 3.7-lb/d reduction in milk yield compared with TN-TN. Postpartum HS was associated with increases of 4.4-percentage points in incidence of retained placenta, 18.1-percentage points in incidence of metritis, and 2.0-percentage points in incidence of mastitis, and a reduction in 5.3-percentage points in pregnancy at first AI, and an increase in 4.5-percentage points in pregnancy loss compared with Post TN. In cows, exposure to HS during the Pre, Post, or Pre and Post was associated with a 5.3-lb/d reduction in milk yield when compared with TN-TN. Post HS was associated with an increased incidence of retained placenta of 5.8-percentage points within Pre HS cows, whereas no difference was found within Pre TN cows. Metritis was increased by 6.3-percentage points in Post HS cows. For pregnancy per AI, Post HS was associated with reduced pregnancy of 10.6-percentage points within Pre TN cows, whereas no difference was found within Pre HS cows. Removal from the herd increased in cows exposed to HS during the Pre or Post, or Pre and Post. These data suggest that early postpartum HS is associated with performance losses to a greater extent than prepartum HS and that heifers and cows are vulnerable to losses associated with exposure to HS during the transition period.

Introduction

Genetic selection combined with improved management of dairy cows has resulted in an increment of more than 400% in individual cow milk yield over the last century. As production increases, a parallel increase in DM intake occurs to support the nutritional needs for milk synthesis, which influences the metabolic rate and results in additional heat production. More productive cows must dissipate additional heat, which makes them less capable of maintaining body temperature and more susceptible to heat stress (HS). In U.S. dairies, annual economic losses associated with HS are estimated to

be close to \$900 million. Hence, attention to the detrimental impacts of HS on cow performance and farm profitability has been extensively evaluated, including periods when cows are more challenged, such as during the transition period.

Throughout the prepartum period, cows undergo a process of recovery. Heat stress impairs proper recovering of the mammary gland due to the reduced cell functioning, consequently compromising productive performance in the subsequent lactation. Yet, the interaction between HS exposure during the prepartum and postpartum periods on productive performance remains uncertain. Peripartum diseases, such as metritis and mastitis, are highly prevalent during early postpartum with significant consequences to productive and reproductive performance, and survival of dairy cows. Adequate function of the immune system is essential to protect the uterus and mammary gland tissue from bacteria. It is known that providing cooling to cows increases the capacity of the immune system to fight bacteria. In fact, the literature reported that cows exposed to HS during the prepartum period have greater incidence of retained placenta, mastitis, and respiratory problems. Heat stress also has serious negative impacts on reproduction in dairy cows, likely because of the negative impacts of high body temperature on the reproductive physiology.

The literature has limited information regarding the topics of the timing during lactation when HS has its greatest impacts, and about the combined impacts of HS in late gestation and early lactation on production, health, reproduction, and survival in dairy cows. Hence, the objectives were to determine the association between timing of HS exposure during the transition period on production, health, reproduction, and survival during the first 90 days postpartum in Holstein cows. The combined exposure to HS during the pre and postpartum periods was thought to have the greatest impact on cow performance compared to HS only prepartum or only postpartum, and the latter two were expected to be detrimental to cows compared with no HS exposure. Furthermore, the impacts of HS were expected to affect heifers and cows.

Experimental Procedures

Study Design

Data from two commercial herds located in central California were collected for this retrospective cohort study. Cows were classified based on temperature humidity index (THI) exposure during the last 28 days of gestation (Pre) and the first 28 days postpartum (Post) and considered as exposed to HS when the average THI in the pre and postpartum periods were greater than 68. To address the exposure to HS during either in the Pre or Post or to both periods combined, only cows that calved in April, June, July, and September of each year (2012, 2013, and 2014) remained in the study. A total of 5,722 Holstein cows (2,324 heifers and 3,397 cows) were used for the study and categorized into EC groups based on THI exposure during the Pre and Post as HS or thermoneutral (TN), resulting in four different groups; TN-TN ($n = 1,040$; mean THI Pre = 57.9 and Post = 63.7), TN-HS ($n = 1,214$; mean THI Pre = 68.2 and Post = 72.0), HS-TN ($n = 1,917$; mean THI Pre = 71.4 and Post = 65.6), and HS-HS ($n = 1,551$; mean THI Pre = 72.2 and Post = 72.7).

Data Collection

Weather data were obtained from the station located at the Visalia Municipal Airport, Visalia, CA, approximately 8 miles from both farms. The data collected included the

daily average dry bulb temperature and the relative humidity, which were used to calculate the THI as previously described. Health, production, reproduction, and survival data in the first 90 days postpartum were retrieved from the dairy management software Dairy Comp 305. Production was measured for the first 3 months postpartum, and it was assumed that milk yield observed on that day represented the average production of the respective month postpartum. The cumulative milk yield by 90 days postpartum was computed by summing the daily milk production in the first 90 days postpartum or until the cow left the herd if before 90 days postpartum. In herd 1, cows were housed in dry lots with shades in the central area of the pen and over the feed bunk. Dry composted manure was used as bedding material and the shaded area was raked once daily. In the feed lane, soaker lines with nozzles were placed above the stanchions, and were activated for 1 min each 6 min when the ambient temperature reached 71.6 °F. In herd 2, lactating cows were housed in free-stall barns with sand as the bedding material. Dry cows were also housed in free-stall barns, but during the prepartum period cows were housed in dry lot pens until parturition. Barns were equipped with fans in the central area above the beds. In the feed lane, soaker lines with nozzles were placed above the stanchions, and were activated for 1 min every 6 min when the ambient temperature reached 71.6°F.

Reproductive Management

Both herds used the same reproductive management program. Synchronization of estrous cycle was performed by administration of 25 mg of prostaglandin (PGF_{2α}) 14 days apart at 37 and 51 ± 3 days postpartum. Detection of signs of estrus based on removal of chalk from the tail started at 51 ± 3 days postpartum and those in estrus were inseminated on the same day. Cows not inseminated within 11 days after the second dose of PGF_{2α} treatment were enrolled in the Ovsynch-56 timed AI protocol starting on day 62 ± 3 postpartum, such that every cow was programmed to receive their first AI by 75 ± 3 days postpartum. Cows were observed for estrus daily as previously described, and those that returned to estrus were inseminated in the same morning and considered to be nonpregnant to the previous breeding. Pregnancy was diagnosed by transrectal ultrasonography on days 32 ± 3 after AI. The presence of an amniotic vesicle containing an embryo with a heartbeat was used as the criteria to determine pregnancy. Pregnant cows on day 32 after AI were reexamined for pregnancy 4 weeks later, on day 60 of gestation. Pregnancy per AI was calculated by dividing the number of cows diagnosed pregnant at day 32 or 60 after AI by the number of cows receiving AI. Reproductive data were collected only for the first AI.

Statistical Analysis

Heifers and cows were analyzed separately using the same models to facilitate interpretation of the data. Daily or cumulative milk yields were analyzed by ANOVA using the MIXED procedure of SAS. Distribution of residuals and homogeneity of variance were evaluated after fitting the statistical models for continuous data. Risk of individual diseases or morbidity, P/AI, and pregnancy loss were analyzed by logistic regression using the GLIMMIX procedure of SAS. The interval from calving to diagnosis of the first disease event or to removal from the herd were analyzed with Cox's hazard regression model using the PHREG procedure of SAS.

Results and Discussion

Productive Performance

Exposure to HS during the Pre or Post, or Pre and Post was associated with a reduction ($P < 0.05$) in milk yield of 3.7-lb/d and a total of 392 lb in the first 90 days postpartum compared with TN-TN (Table 1). Like heifers, exposure to HS during the Pre or Post, or Pre and Post in cows was associated with a reduction ($P < 0.05$) in milk yield of 2.3-lb/day resulting in a cumulative decrease of 592 lb in the first 90 days postpartum compared with TN-TN (Table 2).

Incidence of Diseases

In heifers, exposure to HS during the Pre or Post, or Pre and Post was associated with increased ($P < 0.05$) incidence of metritis of 12-percentage points compared with TN-TN (Table 1), and postpartum HS exposure was associated ($P = 0.03$) with increased incidence of mastitis (Post TN = 3.3% vs. Post HS = 5.2%). Morbidity affected 54.3% of heifers, and exposure to HS during the Pre, Post, or Pre and Post was associated with an 18-percentage point increase ($P < 0.05$) in morbidity compared with TN-TN (Table 1). In addition, the hazard of morbidity in the first 90 days postpartum increased ($P < 0.001$) 72%, 74%, and 89% for TN-HS, HS-TN, and HS-HS, respectively, compared with TN-TN (Figure 1A). In cows, among cows that experienced Pre HS, those that also experienced Post HS (HS-HS) had greater ($P < 0.05$) incidence of retained placenta compared with HS-TN. Postpartum HS exposure was associated ($P = 0.009$) with increased incidence of metritis (Post TN = 19.7% vs. Post HS = 29.4%) and tended ($P = 0.08$) to be associated with increased incidence of mastitis (Post TN = 8.0% vs. Post HS = 9.9%). Morbidity affected 33.8% of cows, and Postpartum HS exposure tended ($P = 0.07$) to be associated with increased morbidity (Post TN = 41.2 vs. Post HS = 44.7 ± 2.3 ; Table 2).

Reproductive Performance

Prepartum HS exposure was not associated with pregnancy at first AI in heifers. Postpartum HS exposure tended ($P = 0.07$) to be associated with reduced P/AI on day 32 after the first AI (Post TN = 42.1% vs. Post HS = 38.1%) and was associated ($P = 0.01$) with reduced P/AI on day 60 after first AI (Post TN = 38.7% vs. Post HS = 33.4%). This is because heifers exposed to Post HS had increased ($P = 0.04$) pregnancy loss (Post TN = 7.8% vs. Post HS = 12.3%; Table 1). In cows, exposure to Post HS was associated with depressed P/AI at 32 and 60 days after first AI, but the impact was greater in cows exposed to Pre TN. There was no association between HS exposure and loss of pregnancy in cows.

Survival

In heifers, prepartum HS exposure was associated ($P = 0.04$) with increased removal from the herd (Pre TN = 6.5% vs. Pre HS = 8.8%; Table 1). Likewise, Post HS exposure was associated ($P = 0.001$) with increased removal from the herd (Post TN = 5.7% vs. Post HS = 9.6%). The hazards of leaving the herd by 90 days postpartum were 125%, 81%, and 163% greater ($P < 0.05$) for TN-HS, HS-TN, and HS-HS, respectively, compared with TN-TN (Figure 1C). In cows, a tendency for an interaction ($P = 0.09$) between Pre and Post exposure to HS was observed for removal from the herd. Exposure to HS during the Pre, Post, or Pre and Post were associated with greater ($P < 0.05$) removal from the herd by 5-percentage points compared with TN-TN (Table 2). In

addition, the hazards of removal from the herd were 60%, 48%, and 59% greater ($P < 0.05$) for TN-HS, HS-TN, and HS-HS, respectively, compared with TN-TN (Figure 1D).

Discussion

This study demonstrates the association between HS exposure throughout the late gestation and the early lactation period with production, risk of diseases, reproduction, and survival in the first 90 days postpartum in Holstein dairy cows. The literature is vast on experiments evaluating the effects of providing evaporative cooling to cows exposed to HS on productive performance; however, limited data exist from controlled experiments evaluating the impact of providing heat abatement on health and survival in dairy cows. The present observational study was designed to attempt to isolate the impacts of exposure to HS at critical times of the lactation cycle, late gestation, and early lactation on postpartum performance. Although an observational study is limited as there are other variables in addition to the variable of interest, our results showed that cows exposed to HS either Pre or Post were associated with impaired production, reproduction, and more likely to develop early lactation diseases. In almost all instances, exposure to HS either Pre or Post or in both periods was associated with depressed production, reproduction, and increased risk of diseases in heifers and cows.

It is well established that reducing HS by providing evaporative cooling during the entire dry period or during lactation benefits the lactation performance in dairy cows, although most of the data from controlled experiments involved cows. Parity is an important factor to be considered because heifers and cows do not always have the same response to dietary and management interventions during the transition period. Heifers are still growing and have nutritional needs to address growth of tissue. Also, heifers undergo extensive mammogenesis during late pregnancy. Thus, it is possible that impacts of exposure to HS might differ between the two parity groups. The present data clearly showed that both heifers and cows suffer when exposed to HS either Pre or Post or in both periods. One of the most common observations in cows that suffer from HS is the reduction of milk yield. Although it has been suggested that early lactation cows suffer less from the detrimental impacts of HS compared with mid and late lactation cows, the present findings reveal that exposure to HS at any time in the transition period was associated with depressed production in both heifers and cows. When exposed to high ambient temperature or high THI, dairy cows are unable to properly thermoregulate and they often respond with reduced DMI, which is thought to explain approximately half of the reduced milk yield. In addition, cows exposed to HS have reduced numbers of mammary epithelial cells resulting in reduced milk yield. Thus, the combined reduction in nutrient supply and direct effects of hyperthermia on the mammary gland likely explains the reduced milk production in cows exposed to HS.

Adequate innate immune function is essential to protect cows against infections. Providing evaporative cooling to cows exposed to HS enhances innate immune cell function, lymphocytic proliferation, and TNF α expression. Likewise, immune cells have less phagocytosis and oxidative burst when exposed to increased temperature. Not only are cows impacted by changes in environmental conditions, the increased temperatures are associated with more pathogens in the environment of the cow, which likely contributes to an increased risk of infectious diseases. Environmental conditions, such as HS, likely trigger a combination of impaired host resistance and

increased pathogen challenge that favors disease. In the present study, exposure to HS at any time in the transition period in heifers and Post HS in cows was associated with increased risk of developing disease postpartum. Hyperthermia is known to impact follicle growth, oocyte quality, early embryo development, endometrial function, and the endocrine milieu in dairy cows. This study clearly showed that exposure to HS, particularly postpartum, depressed P/AI in both heifers and cows and increased the risk of pregnancy loss in heifers. Cows exposed to HS have impaired endocrine pathways that alter the production and secretion of ovarian hormones such as estradiol and inhibin, which affects follicular dominance. Secretion of LH and progesterone are altered by HS, which can impair final maturation and ovulation of the pre-ovulatory follicle and affect establishment and maintenance of pregnancy. Development of the pre-ovulatory follicle begins months before ovulation, and insults such as HS or diseases can have long-lasting effects on reproduction. Oocytes can be damaged by HS as early as 105 days before ovulation. Inflammatory diseases that affect cows in early lactation reduce morula and conceptus quality and induce damages to the conceptus that carry on during later stages of pregnancy. Thus, it is possible that direct insults caused by HS and the associated increased risk of diseases observed in the present study explain the reduced P/AI and increased risk of pregnancy loss.

Conclusion

Exposure to heat stress either in late gestation or in early lactation was associated with reduced milk production in the first 90 days of lactation. Nevertheless, early postpartum heat stress was associated with performance losses to a greater extent than prepartum HS, mainly because of more adverse health and reproduction responses. Exposure to heat stress postpartum increased morbidity and reduced pregnancy per AI, both of which are known to impact survivability of cows in dairy herds. In fact, heifers and cows exposed to heat stress had increased hazard of leaving the herd in early lactation. Based on the current findings, it is possible that heat abatement pre and postpartum would improve performance but suggests that priority should be given to cows in early lactation. Controlled experiments evaluating the effect of heat abatement pre and postpartum on health and reproductive performance warrant further research.

Table 1. Association between environmental condition (EC) exposure during the transition period and performance during the first 90 d postpartum in Holstein heifers

Item	EC ¹				P-value ²		Pre × Post
	TN-TN	TN-HS	HS-TN	HS-HS	Pre	Post	
Yields of milk							
lb/d	69.7 ^a	66.0 ^b	66.6 ^b	65.6 ^b	< 0.001	< 0.001	< 0.001
Cumulative 90 d, lb	6,202 ^a	5,810 ^b	5,918 ^b	5,793 ^b	< 0.001	< 0.001	< 0.001
Disease, ³ %							
Retained placenta	9.1	14.1	11.2	12.0	0.90	0.14	0.29
Metritis	36.3 ^a	55.5 ^b	48.0 ^b	53.0 ^b	0.07	< 0.001	0.007
Mastitis	2.6	5.0	4.0	5.5	0.24	0.03	0.47
Morbidity ⁴	40.4 ^a	58.5 ^b	58.1 ^b	61.4 ^b	< 0.001	< 0.001	0.0006
Pregnant at 1st AI, %							
day 32	43.6	40.1	40.7	36.2	0.12	0.07	0.79
day 60	40.3	34.8	37.2	32.1	0.16	0.01	0.96
Pregnancy loss	7.2	13.2	8.5	11.5	0.95	0.04	0.50
Removed by 90 d, ³ %	4.1	8.9	7.2	10.3	0.04	0.001	0.24

¹ Environmental condition was categorized based on temperature humidity index (THI) exposure during the pre and postpartum periods as TN-TN (mean THI Pre = 57.9 and Post = 63.7), TN-HS (mean THI Pre = 68.2 and Post = 72.0), HS-TN (mean THI Pre = 71.4 and Post = 65.6), or HS-HS (mean THI Pre = 72.2 and Post = 72.7). TN = thermoneutral, HS = heat stress.

² Pre = effect of prepartum exposure to heat stress (TN-TN + TN-HS vs. HS-TN + HS-HS); Post = effect of postpartum exposure to heat stress (TN-TN + HS-TN vs. TN-HS + HS-HS); Pre × Post = interaction between the effect of prepartum and postpartum exposure to heat stress (TN-TN + HS-HS vs. TN-HS + HS-TN).

³ Evaluated in the first 90 d postpartum.

⁴ Morbidity defined as cows having at least one of the following diseases: retained placenta, metritis, mastitis, lameness, displaced abomasum, or pneumonia.

Table 2. Association between environmental condition (EC) exposure during the transition period and performance during the first 90 d postpartum in Holstein cows

Item	EC ¹				P-value ²		Pre × Post
	TN-TN	TN-HS	HS-TN	HS-HS	Pre	Post	
Yields of milk							
lb/d	94.8 ^a	89.7 ^b	89.5 ^b	88.0 ^b	< 0.001	< 0.001	< 0.001
Cumulative 90 d, lb	8,384 ^a	7,792 ^b	7,839 ^b	7,643 ^b	< 0.001	< 0.001	< 0.001
Disease, ³ %							
Retained placenta	14.1	11.5	8.2 ^a	14.0 ^b	0.27	0.28	0.02
Metritis	25.4	29.4	19.7	27.5	0.08	0.009	0.34
Mastitis	7.5	10.7	8.5	9.1	0.87	0.08	0.22
Morbidity ⁴	39.9	43.2	42.6	46.2	0.13	0.07	0.93
Pregnant at 1st AI, %							
day 32	36.2 ^a	23.2 ^b	33.2 ^a	30.9 ^a	0.13	< 0.001	0.002
day 60	30.4 ^a	19.8 ^b	28.2 ^a	25.7 ^a	0.20	< 0.001	0.01
Pregnancy loss	15.5	12.0	13.8	14.3	0.88	0.55	0.44
Removed by 90 d, ³ %	8.5	13.2	12.4	13.2	0.09	0.02	0.09

¹ Environmental condition was categorized based on temperature humidity index (THI) exposure during the pre and postpartum periods as TN-TN (mean THI Pre = 57.9 and Post = 63.7), TN-HS (mean THI Pre = 68.2 and Post = 72.0), HS-TN (mean THI Pre = 71.4 and Post = 65.6), or HS-HS (mean THI Pre = 72.2 and Post = 72.7). TN = thermoneutral, HS = heat stress.

² Pre = effect of prepartum exposure to heat stress (TN-TN + TN-HS vs. HS-TN + HS-HS); Post = effect of postpartum exposure to heat stress (TN-TN + HS-TN vs. TN-HS + HS-HS); Pre × Post = interaction between the effect of prepartum and postpartum exposure to heat stress (TN-TN + HS-HS vs. TN-HS + HS-TN).

³ Evaluated in the first 90 d postpartum.

⁴ Morbidity defined as cows having at least one of the following diseases: retained placenta, metritis, mastitis, lameness, displaced abomasum, or pneumonia.

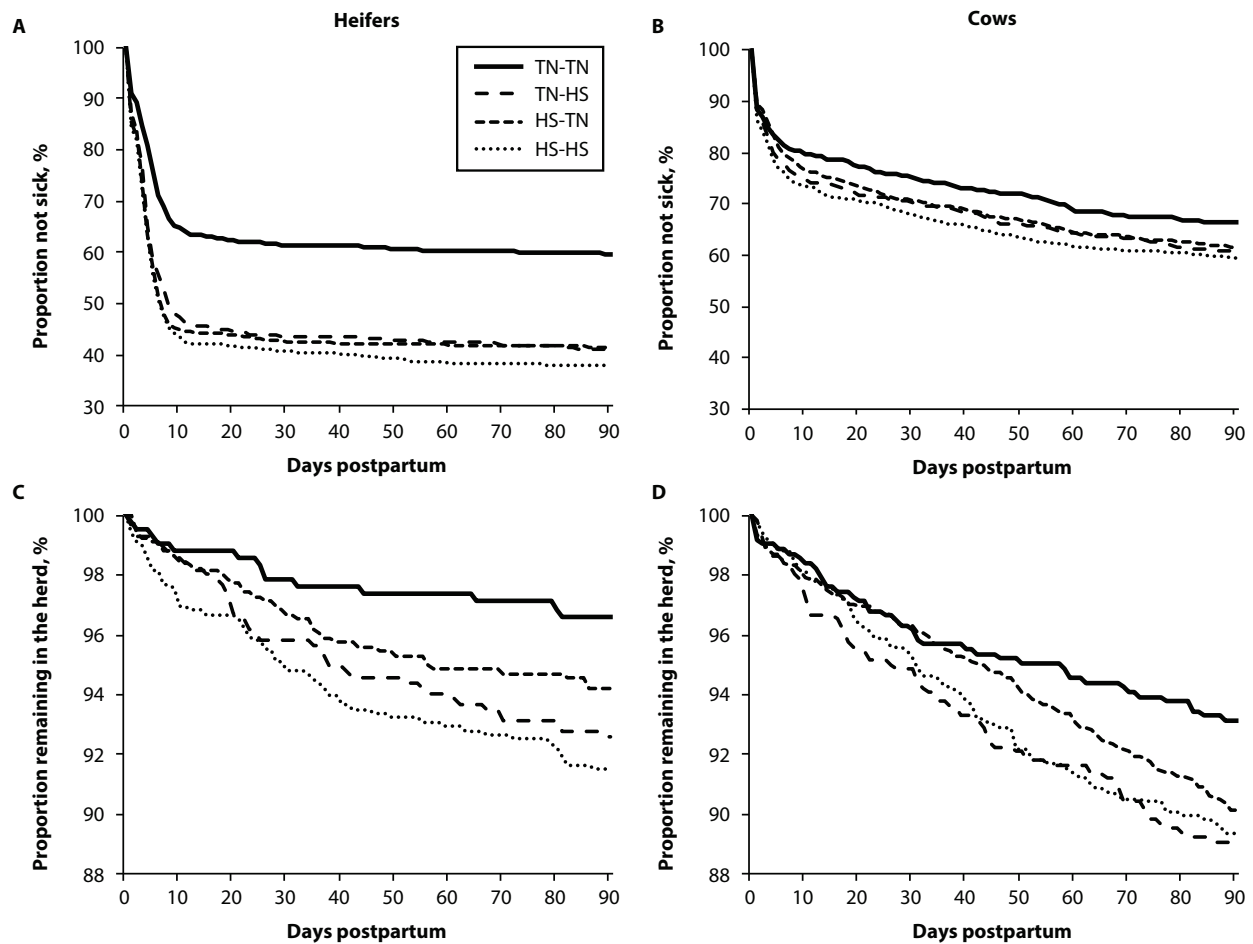


Figure 1. Survival curves for days to diagnosis of disease in Holstein heifers (A) and (B) cows, and survival curves for days to removal from the herd in Holstein heifers (C) and cows (D) in the first 90 d postpartum classified based on environmental condition (EC) exposure. TN = thermoneutral. HS = heat stress.