

DAILY TEMPERATURE VARIATIONS AND ITS IMPACT ON HIGH ALTITUDE DAIRY PRODUCTION

VARIACIONES DIARIAS DE TEMPERATURA Y SU IMPACTO EN LA PRODUCCIÓN DE LECHE EN ALTURA

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Recibido: 10/06/2022

Aceptado: 05/09/2022

ABSTRACT. Extensive dairy productions are exposed to weather variations, and when the temperature and humidity are extreme, animals can experience thermal stress. The aim of this work was to evaluate the impact of a daily variation on temperature-humidity index (THI) on circulating cortisol levels, milk yield and composition of dairy cows. Ten Holstein cows in the first third of lactation were evaluated, during morning and afternoon milking, for milk yield, milk composition and blood cortisol. Climatic variables were recorded daily. During the experimental period, the daily temperature variations were around 15°C (range between maximum and minimum), however THI indicated animals were in a thermoneutral zone. Milk composition and blood cortisol concentrations were not different between morning and afternoon milking and blood cortisol concentration had a weak positive correlation with milk yield and negative with milk protein, fat and non-fat solids content. In conclusion, Holstein cows located in regions of high altitude do not show a significant level of relationship between blood cortisol and production and milk components, which may indicate the adaptation of these animals to this environmental condition.

KEYWORDS: cortisol, milk production, temperature-humidity index.

RESUMEN. Los sistemas ganaderos extensivos están expuestos a las variaciones climáticas, y cuando la temperatura y humedad son extremas, los animales pueden enfrentar estrés térmico. El objetivo de este estudio fue evaluar el impacto de la variación diaria de la temperatura y el índice temperatura y humedad (ITH) sobre las concentraciones sanguíneas de cortisol, producción y composición de leche en vacas Holstein. Diez vacas fueron seleccionadas, evaluadas en el primer tercio de lactancia, durante los ordeños de la mañana y tarde, para analizar concentraciones sanguíneas de cortisol, producción y composición de leche. Las variables climáticas fueron reportadas diariamente. Durante el periodo experimental, la variación diaria de temperatura estuvo al redor de 15°C (rango entre máxima y mínima), pero el ITH indicó que los animales estaban en una zona termoneutral. La composición de la leche y la concentración sanguínea de cortisol no fue diferente entre los ordeños de la mañana y tarde y el cortisol sanguíneo tuvo una correlación baja, positiva con producción de leche y negativa con la concentración de proteína, grasa y sólidos de la leche. En conclusión, vacas Holstein ubicadas en regiones de alta altitud no evidencian un nivel significativo de relación entre cortisol sanguíneo con la producción y componentes de la leche, lo que puede indicar adaptación de estos animales a esta condición ambiental.

PALABRAS CLAVE: cortisol, índice de temperatura y humedad, producción de leche.

INTRODUCCIÓN

Extreme weather conditions play a significant adverse effect in production systems, especially on large-scale productions such as livestock. The increase in temperature and humidity index (THI) is the climatic condition that most induces stress (Kekana, Nherera-Chokuda, Muya, Manyama, & Lehloenya, 2018). Short-term heat stress in dairy cows requires energy to dissipate it, which results in an increase in the basal temperature and a decrease in dry matter intake, reducing the lactation performance during 24 to 48 hours after the thermal stress (Gantner et al., 2015; Gaughan et al., 2010; Pragna et al., 2017).

High altitude regions have a specific characteristic, which is a significant temperature variation within 24 hours, with high temperatures in the afternoon compared with very low temperatures in the morning. Physiological processes are affected by the circadian rhythm, which includes the eating pattern of dairy cows (Devries & Von Keyserlingk, 2005), as well as, the environmental temperature, reported as an increase in night feeding during warmer periods (Niu & Harvatine, 2018a).

The impact of high temperatures on cattle is reflected in a decreased feed consumption, digestion and absorption of nutrients, increased blood flow to the body periphery and reduced nutrients flow to the udder, affecting milk yield and composition and the immune system (Moretti, Biffani, Chessa, & Bozzi, 2017; Summer, Lora, Formaggioni, & Gottardo, 2019; Tao & Dahl, 2013). Meanwhile, the hypothalamic-pituitary-adrenal axis (HPA) promotes the increase of glucocorticoids in the cortex of the adrenal gland such as cortisol, which contributes to hyperglycemia through stimulation of hepatic gluconeogenesis, inhibition of peripheral glucose utilization and the uptake and metabolism of glucose necessary for milk production (Lefcourt, Bitman, Kahl, & Wood, 1993). Cortisol is released by stressful stimuli, using other metabolic pathways such as proteolysis and lipolysis aiming to regulate the metabolism (Rees, Fischer-Tenhagen, & Heuwieser, 2016), and has a negative effect on milk composition (Fukasawa, Tsukada, Kosako, & Yamada, 2008; Liu et al., 2017).

This study aimed to evaluate the impact of a daily temperature variation on circulating cortisol levels, milk yield and composition of dairy cows on a high-altitude environment.

MATERIALS AND METHODS

This study included ten multiparous Holstein cows in the first third of lactation (54 \pm 33 DIM, 14 Kg/d average production in previous lactation) from a production system owned by La Salle University. The farm is located at the municipality of Sopo, Cundinamarca, Colombia, at 2,587 m of altitude. The evaluations were performed during the dry season, when the average temperature was 13.3 °C (minimum: 2.8 °C; maximum: 23.1 °C); humidity 83.6 % (minimum: 35.4 %; maximum: 98 %); THI: 56 (minimum: 40; maximum: 66) and average precipitation of 116 mm. These parameters were obtained from a weather station (Vantage Pro2, Davis instruments, country) placed at the farm.

Animals were kept in a pasture-based system (mainly *Cenchrus clandestinus*, *Lolium* sp. and *Trifolium pratense*), with regular rotations and supplemented twice a day at milking with a commercial concentrate (19 % CP; 4.4 Mcal/Kg ME in a ration of 1 kg of supplement for each 5 L of milk production), mineral mix supplementation and 2 kg of corn silage (8 % CP; 4.2 Mcal/Kg ME).

Samples were taken in four different days, twice during AM milking (3:00 AM) and twice during PM milking (3:00 PM). On each day milk yield was recorded and milk samples were collected directly from the milking line and immediately refrigerated at 4°C until analyzed. Milk components (protein, fat and non-

fat solids percentage) were measured using an automatic system (LactoStar® milk analyzer, Funke Gerber, country). Blood samples were obtained from the vein-artery coccygeal complex in 5-mL vacuum tubes containing EDTA. After collection, tubes with blood samples were placed on ice and then centrifugated at 1,800 xg for 20 minutes, plasma was harvested and stored at -80°C until further analysis. Blood cortisol was analyzed with a commercial kit (Cortisol AccuBind® ELISA, Monobind Inc, country) in duplicate, using a SQ2800 spectrophotometer (Mindray, MR-96A, country).

The temperature-humidity index (THI) was calculated every 6h, using the equation proposed by Kibler (1964): “ $THI = 1.8T_a - (1 - RH)(T_a - 14.3) + 32$ ”, where: T_a - measured ambient temperature in °C and RH - relative humidity as a fraction of the unit.

Statistical analysis was performed with an ANOVA procedure of SAS 9.3 (SAS Institute Inc., Cary, NC). The fixed effect in the model was the time of sampling (AM or PM), and cows were selected randomly. Least squares mean separation was performed using the t-test. Also, the relation between blood cortisol concentration and milk variables was evaluated with a Spearman correlation test. Statistical significance was declared at $P \leq 0.05$, and tendency as $0.05 \leq P \leq 0.10$.

RESULTS AND DISCUSSION

Minimum and maximum temperature and THI were calculated in the day before each bleeding (Table 1). Results indicated that cows were not experiencing heat stress, according to literature that establish the threshold of THI higher than 72 to indicate heat stress for Holstein breed (Kekana, Nherera-Chokuda, Muya, Manyama, & Lehloenya, 2018). However, it was possible to identify important temperature variations in the same day as its possible to see on day 3, when the minimum temperature was 6.05°C and the maximum temperature 21.41°C, a 15.36°C of variation, which is common to observe in high altitude locations of tropical regions. Additionally, thermal sensation in high altitude areas is greater. Temperature variation leads animals to experience stress and requires a greater energy investment for homeostasis, alters the rhythm of hormonal secretion, modifies the metabolic rates, decreases feed intake, and induces changes in behavior, which limits their productive capacity (Leliveld et al., 2022).

Table 1. THI, minimum and maximum temperature during the experimental period calculated each 6 h in de previous day of sampling.

Day	Hour interval	THI	Temperature, °C	
			min	max
1	12 am – 6 am	51.0	9.86	11.2
	7 am – 11 am	56.9	10.4	17.3
	12 pm – 6 pm	60.6	12.7	19.3
	7 pm – 11 pm	52.8	10.6	12.4
2	12 am – 6 am	51.8	9.89	11.5
	7 am – 11 am	56.8	11.4	16.1
	12 pm – 6 pm	61.7	15.8	17.6
	7 pm – 11 pm	53.8	10.9	14.3
3	12 am – 6 am	44.5	6.05	8.40
	7 am – 11 am	56.4	7.14	19.5
	12 pm – 6 pm	65.4	17.8	21.4
	7 pm – 11 pm	52.4	7.55	15.3

4	12 am – 6 am	45.7	6.81	8.00
	7 am – 11 am	55.0	6.74	19.8
	12 pm – 6 pm	65.8	18.0	22.1
	7 pm – 11 pm	53.4	7.68	16.3

As expected, AM milk yield was higher than PM milk ($P = 0.004$ Table 2). It could be explained due to the availability of blood glucose, lower activity during the night shift and circadian rhythm, which has important elements closely related to energy metabolism regulation in mammals (Peek, Ramsey, Marcheva, & Bass, 2012; Teng et al., 2021). Regarding milk components, there was no difference between AM and PM milking, as well as blood cortisol concentrations for all parameters ($P > 0.05$; Table 2). It's important to highlight that blood cortisol concentration up to 10 ng/mL is considered physiological to dairy cows (Mormède et al., 2007), which indicates all evaluated animals were not exposed to a stressful situation.

It should be considered that the circadian rhythm could be related to a higher cortisol concentration during the morning, since it is necessary to have blood glucose available to supply the energy requirements for basal and metabolic activity (Kendall, Tucker, Dalley, Clark, & Webster, 2008; Niu & Harvatine, 2018b). However, the existence of an adaptability response in cattle can support the lack of cortisol variation observed in this study, which can be explained at a metabolic and molecular level, involving adaptations in the heart, kidney, liver, lung, skeletal muscle, and spleen (Kong et al., 2021; Ma et al., 2022). Taken together, this information suggest that the dairy cows evaluated in this study were adapted to this specific high altitude environmental condition.

Table 2. Milk yield, components and blood cortisol observed at the AM milking (3:00 AM) and at PM milking (3:00 PM).

Variable	AM milking	PM milking	<i>P</i> value
Milk yield (L/d)	10.6 ± 0.67	9.00 ± 0.67	0.004
Milk protein (%)	2.97 ± 0.08	2.88 ± 0.08	0.173
Milk fat (%)	3.76 ± 0.28	3.41 ± 0.28	0.293
Non-fat solids (%)	8.81 ± 0.21	8.68 ± 0.21	0.399
Blood cortisol (ng/mL)	2.70 ± 0.27	2.42 ± 0.27	0.168

In high altitude regions, it is common to see great temperature variation on the same day, which can affect milk yield, fat and protein components. Also, Negrão & Marnet (2006) and Kadzere, Murphy, Silanikove, & Maltz (2002) mention that increased milk production is related to higher cortisol concentration, (as expected in a physiological condition), due to the metabolic requirement. The understanding of cattle adaptation to climate changes is multi-dimensional, dairy cows exposed to heat stress decrease dry matter intake and consequently reduce milk production, rather with a delay (24 to 48 h) (Gauly & Ammer, 2020). Regarding milk components, literature reports have higher variation, however, protein and fat are the most negatively affected in Holstein cows under moderate and severe heat stress (Smith, Smith, Rude, & Ward, 2013).

Spearman correlation test (Table 3) showed that blood cortisol had a weak positive correlation with milk yield, but a negative correlation with milk components. Depending on the feeding regimen, which is highly impacted by environmental characteristics in grazing systems, the

mammary gland will adapt milk yield and composition, in cases of stress due to an increase in THI, cortisol level will induce insulin resistance, dropping glucose availability, consequently, solid concentration in milk is decreased (Hill & Wall, 2014, 2017; Weber et al., 2016).

Table 3. Spearman correlations between the evaluated parameters (cortisol, milk yield, milk protein, milk fat and non-fat solids).

	Milk yield, L/d	Milk protein, %	Milk fat, %	Non-fat solids, %	Blood cortisol, ng/mL
Milk yield, L/d	*****	0.028	-0.047	-0.010	0.175
Milk protein, %	0.028	*****	0.442	0.964	-0.273
Milk fat, %	-0.047	0.442	*****	0.346	-0.032
Non-fat solids, %	-0.010	0.964	0.346	*****	-0.271
Blood cortisol, ng/mL	0.175	-0.273	-0.032	-0.271	*****

This study did not demonstrate the impact of high daily temperature variations on blood cortisol, and consequently its effects on milk yield and composition. Some aspects to be considered could be the THI, which did not indicate thermal stress on these animals; the adaptation that the Holstein breed undergoes to high-altitude environmental conditions; the experimental design with a greater subset of animal, regarding the sampling time points with different days; are some examples.

Blood cortisol is a useful tool to determine stress of dairy cows exposed to thermal stress, however there are limitations when evaluating this hormone, as its physiological high variation can affect results (Trevisi & Bertoni, 2009). Therefore, other stress biomarkers could be evaluated simultaneously to correlate stress levels in future investigations, such as progesterone, plasma albumin, urea, globulin, haptoglobin, fibrinogen, the accumulated cell volume, leukocyte count, thermal shock proteins, concentrations of β -hydroxybutyrate and reduction in the groups containing short and medium chain fatty acids (Liu et al., 2017; Sporer et al., 2008).

CONCLUSION

Even though the daily temperature variations were high, the temperature-humidity index did not indicate animals were under thermal stress. The milk yield was greater in AM milking; however, milk composition and blood cortisol concentrations were not different between morning and afternoon milking. Also, blood cortisol concentration had a weak positive correlation with milk yield and a weak negative correlation with milk protein, fat and non-fat solids content, indicating that the evaluated Holstein cows may be adapted to high altitude environmental conditions.

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