Theoretical and Applied Veterinary Medicine

ISSN 2663-1156 (print)
ISSN 2663-1164 (online)
Theoretical and Applied Veterinary Medicine, 12(4), 3–8
doi: 10.32819/2024.12016

Original research

Impact of heat stress on blood serum cortisol level in dairy cows

R. V. Mylostyvyi*, M. Wrzecińska, M. Samardžija***, B. V. Gutyj****, V. H. Yefimov*, P. M. Skliarov*, M. O. Lieshchova***
**Dnipro State Agrarian and Economic University, Dnipro, Ukraine*
***West Pomeranian University of Technology, Szczecin, Poland*
****University of Zagreb, Zagreb, Croatia*
*****Stepan Gzhytskyi National University of Veterinary Medicine and Biotechnologies, Lviv, Ukraine*

Received: 29 July 2024
Revised: 23 September 2024
Accepted: 18 November 2024

Dnipro State Agrarian and Economic University, Serhii Efremov Str., 25, 49600, Dnipro, Ukraine

West Pomeranian University of Technology, al. Piastów 17, Szczecin, 70-310, Poland

University of Zagreb, Trg Republike Hrvatske, 14, Zagreb, HR-10000, Croatia

Stepan Gzhytskyi National University of Veterinary Medicine and Biotechnologies Lviv, Pekarska Str., 50, Lviv, 79010, Ukraine

Tel.: +38-097-280-88-19
E-mail: *mylostyvyi.r.v@dsau.dp.ua*

#**Corresponding author:**
R. V. Mylostyvyi
mylostyvyi.r.v@dsau.dp.ua

Abstract. Heat stress (HS) is a critical environmental factor that disrupts the hormonal balance of animals by activating and subsequently suppressing the hypothalamic-pituitary-adrenal (HPA) axis. This process alters cortisol secretion, a key stress hormone involved in maintaining homeostasis, regulating metabolism, and facilitating adaptation to adverse conditions. This study investigates the impact of HS on the hormonal responses of Holstein and Brown Swiss dairy cows, with a specific focus on cortisol as a principal biomarker of stress adaptation. The research was conducted under conditions of acute and chronic HS of moderate intensity. Cows were divided into control (CON) and experimental (HYP) groups. Serum cortisol levels were assessed while accounting for the temperature-humidity index (THI), lactation number, days in milk, and energy-corrected milk yield. Cortisol concentrations were measured in blood serum with using an electrochemiluminescence immunoassay method (ECLIA). The findings revealed that cows in the experimental groups experienced significantly higher daily temperatures and THI compared to the control groups. Holstein cows were subjected to chronic HS for 45 days, with a mean THI of 77.5, while Brown Swiss cows experienced acute HS for 5 days, with a mean THI of 77.6. Serum cortisol levels were significantly lower in the experimental groups: 27.25 ± 2.92 nmol/L versus 39.45 ± 3.26 nmol/L (a 30% decrease) in Holstein cows and 11.6 ± 0.54 nmol/L versus 24.5 ± 8.85 nmol/L (a 52% decrease) in Brown Swiss cows ($p < 0.05$), indicating a suppressive effect of HS on the HPA axis. These results highlight the importance of breed-specific differences in stress responses and underscore the role of cortisol as a key indicator of adaptive mechanisms. The observed reduction in cortisol concentrations suggests potential effects of HS on metabolic stability and resilience to environmental stressors.

Keywords: physiological responses; thermal stress; stress adaptation; temperature-humidity index; breed variability; Holstein cows; Brown Swiss cows.

Вплив теплового стресу на рівень кортизолу в сироватці крові молочних корів

Анотація. Тепловий стрес (HS) є критичним екологічним фактором, що порушує гормональний баланс тварин шляхом активації, а згодом супресії гіпоталамо-гіпофізарно-надниркової (HPA) осі. Це змінює секрецію кортизолу – ключового гормону стресу, який відіграє важливу роль у підтриманні гомеостазу, регуляції метаболізму та забезпеченні адаптації до несприятливих умов. У цій статті досліджено вплив HS на гормональні реакції молочних корів порід голштинської та бурої швіцької, з акцентом на кортизол як основний біомаркер стресової адаптації. Дослідження проводили в умовах гострого та хронічного HS помірної інтенсивності. Корови були поділені на контрольні (CON) та дослідні (HYP) групи. При оцінці концентрації кортизолу в сироватці крові враховувалися температурно-вологісний індекс (THI), кількість лактацій, тривалість днів у молоці та базисний удій. Визначення кортизолу здійснювали за допомогою Вміст кортизолу у сироватці крові визначали за допомогою електрохемілюмінесцентного імуноаналізу (ECLIA). Результати показали, що корови дослідних груп зазнали значно вищих середньодобових температур і THI порівняно з контрольними групами. Голштинські корови піддавалися хронічному HS протягом 45 днів із середнім THI 77,5, тоді як бурі швіцькі корови зазнали гострого HS тривалістю 5 днів із середнім THI 77,6. Концентрація кортизолу в сироватці крові була значно нижчою в дослідних групах: 27,25 ± 2,92 проти 39,45 ± 3,26 нмоль/л (зниження на 30 %) у голштинських корів і 11,6 ± 0,54 проти 24,5 ± 8,85 нмоль/л (зниження на 52 %) у бурих швіцьких корів ($p < 0,05$), що вказує на супресивний вплив HS на HPA вісь. Отримані результати підкреслюють важливість породних відмінностей у реакціях на стрес і підтверджують значення кортизолу як ключового індикатора адаптаційних механізмів. Виявлене зниження рівня кортизолу вказує на можливий вплив HS на метаболічну стабільність і стійкість до стресових факторів.

Ключові слова: фізіологічні реакції; термічний стрес; адаптація до стресу; температурно-вологісний індекс; породні відмінності; голштинські корови; бурі швіцькі корови.

Cite this article: Mylostyvyi, R. V., Wrzecińska, M., Samardžija, M., Gutyj, B. V., Yefimov, V. H., Skliarov, P. M., & Lieshchova, M. O. (2024). Impact of heat stress on blood serum cortisol level in dairy cows. *Theoretical and Applied Veterinary Medicine*, 12(4), 3–8. doi: 10.32819/2024.12016

Introduction

Heat stress (HS) is a significant environmental factor affecting dairy cows, particularly during the warmer months of the year. With rising global temperatures, the impact of HS on the productivity and welfare of livestock has become an increasing concern for the dairy industry. HS triggers a series of physiological responses aimed at maintaining homeostasis, which can negatively affect the overall health and milk production of cows. Consequently, HS adversely impacts the physiology of dairy cattle and the profitability of milk production. The increasing number of days with extreme high temperatures highlights the necessity of assessing HS status in animals using appropriate methods (Sejian et al., 2018; Hoffmann et al., 2021).

HS occurs when heat intake exceeds heat loss, causing disruptions in homeostasis and an increase in body temperature (Lemal et al., 2023). These changes induce various reactions in animals, including behavioural, physiological, neuroendocrine, and molecular alterations (Jurkovich et al., 2024). HS particularly affects cows with a high genetic potential for productivity (Martin-Collado et al., 2024), as they produce more metabolic heat due to their higher milk yields. HS can be induced in dairy cows at air temperatures as higher as 25-26 °C.

The temperature-humidity index (THI), which integrates two key environmental parameters – temperature and relative humidity – is widely used to establish the threshold for the onset of HS (Habeeb et al., 2018). However, this environmental index does not account for individual variability among cows, indicating the need for additional physiological markers to confirm the HS status of particular animals.

One of the key physiological indicators of stress is cortisol concentration – a hormone closely associated with the organism's response to stress. In dairy cows, cortisol levels fluctuate in response to acute and chronic stressors, including temperature variations. During acute HS, when cows experience a short-term significant heat load, cortisol levels significantly increase as part of the stress response (Chen et al., 2023). This elevation facilitates the activation of metabolic processes, increased respiration and heart rate, and energy mobilization to sustain physiological functions (Baumgard & Rhoads, 2013).

However, prolonged exposure to high temperatures, or chronic HS, suppresses cortisol secretion, potentially reducing the ability of cows to cope with stress. This suppression may affect metabolism, immune function, and milk production (Polsky & von Keyserlingk, 2017). Chronic stress can deplete the endocrine system and weaken the immune response, diminishing the organism's ability to produce adequate cortisol levels in response to stress (Polsky & von Keyserlingk, 2017; Chen et al., 2024).

The detrimental effect of HS on the immune system is a primary cause of increased susceptibility to infectious diseases. For example, in the case of metritis, a higher incidence of retained placenta has been associated with elevated cortisol and progesterone levels and reduced estrogen levels (Dovolou et al., 2023). Additionally, low cortisol levels after prolonged HS exposure have been linked to increased pro-inflammatory cytokines, depending on the duration of HS exposure (Chen et al., 2018). Furthermore, a reduction in cortisol levels limits the mobilization of energy and nutrients – such as glucose and fatty acids – necessary for the production of lactose and milk fats, thereby negatively affecting milk yield and composition (Polsky & von Keyserlingk, 2017).

The necessity of studying cortisol levels in the blood serum of dairy cows under HS conditions arises from the complexity of confirming HS status in individual animals. Accurately assessing cortisol levels can provide insight into the degree of physiological stress experienced by dairy cows and help identify those most vulnerable to adverse environmental conditions. The aim of the study was to determine a short-term and long-term effects of heat stress in dairy cows using cortisol levels as a biomarker.

Materials and methods

Animals and Experimental Design

The study involved two breeds of dairy cows: Holstein and Brown Swiss. Eighteen Holstein cows (in their second to third lactations) participated in the study, with eight cows assigned to the experimental group (HYP), which was exposed to hyperthermia during the summer (August), and ten cows assigned to the control group (CON), which was maintained under autumn conditions (October). Additionally, sixteen Brown Swiss cows in their second lactation were included in the study, with eight cows in the experimental group (HYP) experiencing heat stress during the summer (June), and eight cows in the control group (CON) kept under thermally comfortable conditions in autumn (October).

Lactation parameters were similar across both groups. For the Holstein cows, the mean days in milk were 141 ± 11.8 (HYP) and 142 ± 9.7 (CON), with average daily milk yields of 39.5 ± 3.61 kg and 39.3 ± 0.87 kg, respectively. In Brown Swiss cows, the daily milk yields were 106.7 ± 19.12 (HYP) and 136.75 ± 19.1 (CON), with no significant differences for this parameter. Their average daily milk yields were 36.4 ± 6.49 kg and 38.1 ± 6.32 kg, respectively.

Data on the milk production of all cows, including daily milk yield per cow (kg) and the percentages of milk fat and protein, were recorded using the DairyComp 305 herd management system throughout the experimental period. The calculation of the fat-corrected milk (FCM) yield was standardized to a baseline fat content of 3.4%. The following equation applied to present milk yield with aforementioned fat level:

$$\text{FCM (kg)} = \text{Milk Yield (kg)} \times (0.4+0.15\times\text{Fat (\%)})$$

In this equation, FCM (kg) represents the fat-corrected milk yield adjusted to a standardized fat content of 3.4%. "Milk Yield (kg)" denotes the actual daily milk yield, while "Fat (%)" indicates the measured percentage of fat in the milk sample. This method ensured a consistent evaluation of milk production across cows with varying milk fat content.

Housing Conditions

The cows were housed on two commercial dairy farms in the central region of Ukraine in naturally ventilated facilities and kept untethered throughout the year. Sand was used as bedding in the stalls to promote hygiene and enhance animal comfort. All groups received the same total mixed ration (TMR) in each season, balanced for essential nutrients.

Their year-round feed mix was based on maize silage and was formulated according to the recommendations of the National Research Council (NRC, 2001). The diet included high-quality ingredients such as barley, rye, maize grain, maize silage, lucerne silage, grass hay, and wheat straw. Additionally, rapeseed meal, sunflower meal, soybean meal, dried beet pulp, and mineral-vitamin supplements were provided. All ingredients were thoroughly mixed in specialized mixers equipped with electronic scales, and both feeding frequency and ration size were managed by computer software.

The cows had free access to feeders and water troughs, ensuring the consistent availability of feed and water.

Assessment of Climatic Conditions

To determine the climatic parameters and the degree of heat stress in the cows, the temperature-humidity index (THI) was applied, calculated according to Kibler (1964). Data on ambient temperature (°C) and relative humidity (%) were obtained from the nearest meteorological station, located approximately 20 kilometres from the studied dairy farms, and made publicly available via the official website of the Ukrainian Hydrometeorological Centre. The analysis involved paired measurements of temperature and humidity (eight recordings per day at three-hour intervals), from which the daily mean values were calculated during the study period.

Table 1 – Climatic conditions during the study period (Mean ± SE)

Breed (group)	Air temperature, °C	Relative humidity, %	THI, units	Duration > THI 72, days
Holstein (HYP)	28.5 ± 2.1	63 ± 3.5	77.5 ± 0.9	45
Holstein (CON)	20.4 ± 1.8	68 ± 2.7	63.0 ± 0.8	0
Brown Swiss (HYP)	29.1 ± 2.3	62 ± 4.1	77.6 ± 1.1	5
Brown Swiss (CON)	17.9 ± 2.0	69 ± 3.0	66.0 ± 0.7	0

Notes: Duration > THI 72, days – continuous number of days where THI exceeded 72.

Given that 72 units is considered the THI threshold at which most dairy cows begin to experience heat stress (HS), it was inferred that the experimental groups might have been subjected to mild to moderate heat stress. This aligns with the commonly accepted classification: mild stress (72-79), moderate stress (79–89), and severe stress (>89).

In the Holstein HYP group, the THI exceeded 72 for 45 days, with a mean value of 77.5, while in the CON group, it remained at 63. In the Brown Swiss HYP group, a five-day "heat wave" raised the THI to 77-82 (mean 77.6), whereas in the CON group, the THI remained below 68 throughout the observation period.

Hence, the study covered periods during which the HYP groups were subjected to both acute (Brown Swiss) and prolonged (chronic) heat stress (Holstein).

Blood Sampling and Sample Preparation

Blood sampling was performed during the hottest part of the day (13:00–14:00), when the cows were restrained near the feed alley after milking. A total of 10 ml of blood was drawn from the jugular vein of each cow into sterile tubes without anticoagulants. The samples were immediately cooled and subsequently centrifuged at 3000×g for 20 minutes. The serum supernatant was then transferred into 1.5-ml microtubes (Eppendorf AG, Germany) for further analysis.

Determination of Cortisol

The concentration of cortisol was measured using an immunochemical assay with electrochemiluminescence detection (ECLIA) on a Cobas 6000 analyser (module e 601), employing standard reagent kits (Hoffmann-La Roche Ltd, Switzerland).

Ethical Approval

The study was conducted in accordance with the principles of the Declaration of Helsinki and approved by the Commission on Bioethics of the Dnipro State Agrarian and Economic University (protocol No. 5 dated 29 May 2018).

Statistical Analysis

Initially, the data were presented as mean values (Mean) and the standard error of the mean (SE). Since the distribution of most indicators did not meet normality criteria, non-parametric statistical methods were used for further analysis. Statistical processing was performed using STATISTICA 12 (StatSoft, Inc., Tulsa, OK, USA). Significant differences between samples were determined using the Mann–Whitney U test, with a probability level of $p < 0.05$ considered statistically significant.

Results

The results of present study have showed that dairy cows in the experimental groups (HYP) of both breeds experienced significantly higher average daily temperatures, relative humidity, and temperature-humidity index (THI) compared to the control groups (CON) (see Table 1). Holstein cows in the HYP group were exposed to an average daily temperature 8.1 °C higher than the control group and an average THI exceeding the threshold value of 72 units for 45 days, indicating prolonged chronic heat stress. In contrast, Brown Swiss cows in the HYP group experienced an average daily temperature 11.2 °C higher than the control group and an average THI ranging between 77–82 units over 5 days, reflecting acute heat stress. Relative humidity in the experimental groups was slightly lower (by 5–6%) compared to the control groups, although the difference was less pronounced. The control groups of both breeds did not experience heat stress, as their THI remained below 72 units throughout the observation period.

Thus, it can be concluded that experimental animals were exposed to moderate-intensity acute and chronic heat stress, which warrants further analysis of their physiological responses and productive parameters.

Milk production parameters and daily yields did not differ significantly between the experimental (HYP) and control (CON) groups for either breed (see Table 2). In Holstein cows, the mean number of lactation days in the HYP group was 0.7% higher than that of the control group, and daily milk yield was 0.5% higher; however, these differences were not statistically significant ($p > 0.05$). Similarly, in Brown Swiss cows, the lactation period in the HYP group was 22.6% shorter than that of the control group, and daily milk yield was 4.8% lower, but these differences were also not statistically significant ($p > 0.05$), indicating that lactation length did not significantly affect daily milk yields.

The analysis further revealed that serum cortisol concentrations significantly differed between the experimental (HYP) and control (CON) groups for both breeds (see Table 3). In Holstein cows, the mean cortisol level in the HYP group was 30% lower than that of the control group, while in Brown Swiss cows, the cortisol level in the HYP group was 52% lower than that of the control group. All measured cortisol values remained within the reference range (5–50 nmol/L), suggesting that moderate heat stress influenced the hormonal status of the cows without exceeding physiological norms.

Table 2 – Lactation parameters and daily milk yields (Mean ± SE)

Breed (group)	n (animals)	DIM, days	Daily yield, kg	p-value
Holstein (HYP)	8	141 ± 11.8	39.5 ± 3.61	–
Holstein (CON)	10	142 ± 9.7	39.3 ± 0.87	>0.05
Brown Swiss (HYP)	8	106.7 ± 19.12	36.36 ± 6.49	–
Brown Swiss (CON)	8	136.75 ± 19.1	38.10 ± 6.32	>0.05

Notes: p-value – level of statistical significance between groups (HYP vs CON).

Table 3 – Serum cortisol concentrations in cows of different groups (Mean ± SE)

Breed (group)	Cortisol, nmol/L	Reference range, nmol/L	p-value (HYP vs CON)
Holstein (HYP)	27.25 ± 2.92	5-50	<0.05
Holstein (CON)	39.45 ± 3.26	5-50	–
Brown Swiss (HYP)	11.6 ± 0.54	5-50	<0.05
Brown Swiss (CON)	24.5 ± 8.85	5-50	–

Notes: Reference ranges for cortisol concentration are based on Minton (1994).

Given that lactation parameters and daily milk yields did not differ significantly between groups, it can be inferred that variations in cortisol levels were primarily attributable to heat stress rather than differences in production-related parameters.

In summary, the results indicate that dairy cows in the experimental groups of both breeds were subjected to moderate-intensity heat stress of varying durations. Lactation length and daily milk yields were similar between the experimental and control groups, suggesting that metabolic intensity did not significantly influence the results. Instead, serum cortisol concentrations were significantly lower in the experimental groups, highlighting the impact of heat stress on hormonal status. These findings confirm that the observed differences in cortisol levels were directly influenced by heat stress exposure in dairy cows under hyperthermic conditions.

Discussion

Heat stress is a critical environmental factor that activates the hypothalamic-pituitary-adrenal (HPA) axis in dairy cows, regulating cortisol secretion—the primary stress hormone responsible for enabling physiological adaptation to adverse conditions. Cortisol plays a pivotal role in maintaining homeostasis, mobilizing energy resources, and regulating metabolic processes during stressful situations (Ataallahi et al., 2023). However, prolonged elevations in cortisol concentrations can lead to adverse effects, including reduced appetite, impaired reproductive function, decreased milk production, and weakened immune responses (Baumgard & Rhoads, 2013).

Our results revealed that dairy cows in the experimental groups (HYP) of both breeds experienced a significant reduction in serum cortisol concentrations compared to the control groups (CON). Specifically, Holstein cows in the HYP group exhibited a 30% lower cortisol level, while Brown Swiss cows showed a 52% reduction (see Table 3). This underscores the impact of heat stress in lowering cortisol levels, potentially disrupting cortisol's regulatory role in metabolism and stress adaptation (Polsky & von Keyserlingk, 2017; Chen et al., 2018).

Studies confirm that heat stress alters the hormonal status of animals, leading to changes in gene expression in the mammary glands (Mendonca et al., 2025). These changes are associated with decreased milk production and alterations in milk composition due to the regulation of genes responsible for lactose and milk fat synthesis. Similar findings have been reported in other studies demonstrating that heat stress compromises the function of mammary cells, resulting in reduced milk yield (Ponchon et al., 2017; Hooper et al., 2020). Furthermore, heat stress has been shown to modulate the expression of cortisol receptors (GR), prolactin (PRLR), insulin-like growth factor 1 (IGF1R), and insulin receptors (INSR), directly influencing mammary cell function (Hooper et al., 2021; Ouellet et al., 2021).

Cortisol also plays an essential role in mammary gland development during late gestation and initiates the synthesis of milk components in mammary epithelial cells (Lengi et al., 2022). Its effects are regulated through interactions between glucocorticoid (NR3C1) and mineralocorticoid (NR3C2) receptors, which define stress-adaptive mechanisms. However, elevated cortisol concentrations can stimulate apoptosis in mammary tissue by activating the BAX gene and reducing the number of epithelial cells, as demonstrated in studies on chronic stress in goats (Bomfim et al., 2022).

The duration of heat stress plays a crucial role in the physiological responses of dairy cows, particularly in altering cortisol levels, which reflect adaptive processes. Our study demonstrated that Holstein cows subjected to chronic heat stress for 45 days exhibited a 30% reduction in cortisol concentrations, whereas Brown Swiss cows exposed to acute heat stress for 5 days experienced a 52% decline. These results highlight differences in short- and long-term stress responses, likely linked to breed-specific genetic and physiological characteristics (Chen et al., 2018; Sejian et al., 2024).

Marins et al. (2021) demonstrated that stress duration not only affects cortisol levels but also influences the secretion of pro-inflammatory cytokines, such as TNF-α and IL-10, which regulate immune responses. Acute stress triggers a sharp increase in inflammatory markers and cortisol, facilitating energy mobilization and the maintenance of homeostasis. In contrast, chronic stress can lead to the exhaustion of adaptive mechanisms and immune suppression (Cartwright et al., 2023).

Breed-specific differences in heat stress sensitivity play a vital role in dairy cow adaptation to extreme temperatures. Our study confirmed that Holstein cows, known for their high metabolic activity, exhibited greater sensitivity to prolonged heat stress, as evidenced by a 30% reduction in cortisol levels after 45 days of chronic exposure. Conversely, Brown Swiss cows, characterized by higher thermotolerance, showed a 52% decrease in cortisol concentrations after just 5 days of acute stress. These findings indicate breed-related variations in adaptive mechanisms, potentially linked to genetic origins and physiological traits (Polsky & von Keyserlingk, 2017; Mylostyvyi et al., 2021).

Lemal et al. (2023) emphasized the importance of genes such as HSF1 and NFκB, which provide cellular protection against heat stress-induced damage and support mammary gland functionality. These genes regulate heat shock proteins (HSPs) and inflammatory pathways, reinforcing their role in thermotolerance.

Despite the findings, uncertainties remain regarding the long-term effects of chronic stress on productivity and reproductive health, as well as the need for further investigation into genetic adaptation mechanisms (Țogoe & Mincă, 2024). Future research should focus on exploring the genetic and epigenetic mechanisms underlying heat tolerance in dairy cows, particularly the roles of candidate genes such as HSF1, NFκB, and HSPs, which regulate heat shock proteins and inflammatory processes (Lemal et al., 2023). Investigating cortisol interactions with other hormones, including prolactin, insulin, and IGF-1, is crucial for enhancing our understanding of endocrine regulation in stress adaptation (Hooper et al., 2021; Ponchon et al., 2017). The long-term impacts of heat stress on reproductive function and immune responses should also be evaluated, with particular attention to milk production and metabolic stability (Lees et al., 2019; Gupta et al., 2022).

Promising avenues for research include the development of non-invasive stress monitoring methods, such as analyzing cortisol levels in milk and hair, to assess both acute and chronic responses to stressors (Sejian et al., 2022; Kapustka et al., 2024). Additional focus should be given to testing dietary supplements with antioxidant and anti-inflammatory properties as potential strategies for improving heat stress resilience (Marins et al., 2021).

Conclusions

This study highlights the impact of heat stress on the hormonal responses of dairy cows, with a particular focus on cortisol as a key biomarker of stress adaptation. The results demonstrate a significant reduction in serum cortisol concentrations in both Holstein and Brown Swiss cows exposed to moderate acute and chronic heat stress. These findings emphasize the critical role of cortisol in maintaining metabolic stability and adaptive responses while also revealing breed-specific differences in thermotolerance and stress adaptation mechanisms.

Future research should focus on exploring the genetic and epigenetic mechanisms underlying thermotolerance, developing reliable biomarkers for stress monitoring, and implementing strategic approaches to mitigate the effects of heat stress to enhance productivity and animal welfare.

Funding

Present study was not funded with any external financial support.

Acknowledgement

The authors express their sincere gratitude to their respective institutions for providing the resources and support necessary to conduct this research.

Conflict of interests

The authors declare no conflicts of interest.

References

Ataallahi, M., Cheon, S. N., Park, G.-W., Nugrahaeningtyas, E., Jeon, J. H., & Park, K.-H. (2023). Assessment of stress levels in lactating cattle: analyzing cortisol residues in commercial milk products in relation to the temperature-humidity index. Animals, 13(15), 2407.

Baumgard, L. H., & Rhoads, R. P. (2013). Effects of heat stress on postabsorptive metabolism and energetics. Annual Review of Animal Biosciences, 1(1), 311–337.

Bomfim, G. F., Merighe, G. K. F., de Oliveira, S. A., & Negrao, J. A. (2022). Acute and chronic effects of cortisol on milk yield, the expression of key receptors, and apoptosis of mammary epithelial cells in Saanen goats. Journal of Dairy Science, 105(1), 818–830.

Cartwright, S. L., Schmied, J., Karrow, N., & Mallard, B. A. (2023). Impact of heat stress on dairy cattle and selection strategies for thermotolerance: a review. Frontiers in Veterinary Science, 10.

Chen, S., Wang, J., Peng, D., Li, G., Chen, J., & Gu, X. (2018). Exposure to heat-stress environment affects the physiology, circulation levels of cytokines, and microbiome in dairy cows. Scientific Reports, 8(1).

Chen, X., Li, C., Fang, T., Yao, J., & Gu, X. (2024). Effects of heat stress on endocrine, thermoregulatory, and lactation capacity in heat-tolerant and -sensitive dry cows. Frontiers in Veterinary Science, 11.

Chen, X., Shu, H., Sun, F., Yao, J., & Gu, X. (2023). Impact of heat stress on blood, production, and physiological indicators in heat-tolerant and heat-sensitive dairy cows. Animals, 13(16), 2562.

Cobb, T., Hantzopoulou, G.-C., & Narayan, E. (2023). Relationship between wool cortisol, wool quality indices of Australian Merino rams and climatic variables in Tasmania. Frontiers in Animal Science, 4.

Dovolou, E., Giannoulis, T., Nanas, I., & Amiridis, G. S. (2023). Heat stress: a serious disruptor of the reproductive physiology of dairy cows. Animals, 13(11), 1846.

Ghassemi Nejad, J., Park, K.-H., Forghani, F., Lee, H.-G., Lee, J.-S., & Sung, K.-Il. (2019). Measuring hair and blood cortisol in sheep and dairy cattle using RIA and ELISA assay: a comparison. Biological Rhythm Research, 51(6), 887–897.

Gupta, S., Sharma, A., Joy, A., Dunshea, F. R., & Chauhan, S. S. (2022). The Impact of heat stress on immune status of dairy cattle and strategies to ameliorate the negative effects. Animals, 13(1), 107.

Habeeb, A. A., Gad, A. E., & Atta, M. A. (2018). Temperature-humidity indices as indicators to heat stress of climatic conditions with relation to production and reproduction of farm animals. International Journal of Biotechnology and Recent Advances, 1(1), 35–50.

Hoffmann, G., Silpa, M. V., Mylostyvyi, R., & Sejian, V. (2021). Non-invasive methods to quantify the heat stress response in dairy cattle. Climate Change and Livestock Production: Recent Advances and Future Perspectives, 85–98.

Hooper, H. B., dos Santos Silva, P., de Oliveira, S. A., Merighe, G. K. F., Titto, C. G., & Negrão, J. A. (2021). Long-term heat stress at final gestation: physiological and heat shock responses of Saanen goats. International Journal of Biometeorology, 65(12), 2123–2135.

Hooper, H. B., Silva, P. dos S., de Oliveira, S. A., Meringhe, G. K. F., Lacasse, P., & Negrão, J. A. (2020). Effect of heat stress in late gestation on subsequent lactation performance and mammary cell gene expression of Saanen goats. Journal of Dairy Science, 103(2), 1982–1992.

Jurkovich, V., Hejel, P., & Kovács, L. (2024). A review of the effects of stress on dairy cattle behaviour. Animals, 14(14), 2038.

Kapustka, J., Budzyńska, M., Staniszewska, P., Strachecka, A., Staniszewski, A., & Wojtaś, J. (2024). The influence of selected factors on wool cortisol concentration in alpacas (Vicugna pacos). General and Comparative Endocrinology, 350, 114474.

Lees, A. M., Sejian, V., Wallage, A. L., Steel, C. C., Mader, T. L., Lees, J. C., & Gaughan, J. B. (2019). The impact of heat load on cattle. Animals, 9(6), 322.

Lemal, P., May, K., König, S., Schroyen, M., & Gengler, N. (2023). Invited review: from heat stress to disease – immune response and candidate genes involved in cattle thermotolerance. Journal of Dairy Science, 106(7), 4471–4488.

Lengi, A. J., Stewart, J. W., Makris, M., Rhoads, M. L., & Corl, B. A. (2022). Heat stress increases mammary epithelial cells and reduces viable immune cells in milk of dairy cows. Animals, 12(20), 2810.

Marins, T. N., Gao, J., Yang, Q., Binda, R. M., Pessoa, C. M. B., Orellana Rivas, R. M., Garrick, M., Melo, V. H. L. R., Chen, Y.-C., Bernard, J. K., Garcia, M., Chapman, J. D., Kirk, D. J., & Tao, S. (2021). Impact of heat stress and a feed supplement on hormonal and inflammatory responses of dairy cows. Journal of Dairy Science, 104(7), 8276–8289.

Martin-Collado, D., Diaz, C., Ramón, M., Iglesias, A., Milán, M. J., Sánchez-Rodríguez, M., & Carabaño, M. J. (2024). Are farmers motivated to select for heat tolerance? Linking attitudinal factors, perceived climate change impacts, and social trust to farmers' breeding desires. Journal of Dairy Science, 107(4), 2156–2174.

Mendonca, L. C., Carvalho, W. A., Campos, M. M., Souza, G. N., de Oliveira, S. A., Meringhe, G. K. F., & Negrao, J. A. (2025). Heat stress affects milk yield, milk quality, and gene expression profiles in mammary cells of Girolando cows. Journal of Dairy Science, 108(1), 1039–1049.

Minton, J. E. (1994). Function of the hypothalamic-pituitary-adrenal axis and the sympathetic nervous system in models of acute stress in domestic farm animals. Journal of Animal Science, 72(7), 1891–1898.

Mylostyvyi, R., Lesnovskay, O., Karlova, L., Khmeleva, O., Kalinichenko, O., Orishchuk, O., Tsap, S., Begma, N., Cherniy, N., Gutyj, B., & Izhboldina, O. (2021). Brown Swiss cows are more heat resistant than Holstein cows under hot summer conditions of the continental climate of Ukraine. Journal of Animal Behaviour and Biometeorology, 9(4), 1–8.

Ouellet, V., Negrao, J., Skibiel, A. L., Lantigua, V. A., Fabris, T. F., Marrero, M. G., Dado-Senn, B., Laporta, J., & Dahl, G. E. (2021). Endocrine signals altered by heat stress impact dairy cow mammary cellular processes at different stages of the dry period. Animals, 11(2), 563.

Polsky, L., & von Keyserlingk, M. A. G. (2017). Invited review: effects of heat stress on dairy cattle welfare. Journal of Dairy Science, 100(11), 8645–8657.

Ponchon, B., Zhao, X., Ollier, S., & Lacasse, P. (2017). Relationship between glucocorticoids and prolactin during mammary gland stimulation in dairy cows. Journal of Dairy Science, 100(2), 1521–1534.

Sejian, V., Bhatta, R., Gaughan, J. B., Dunshea, F. R., & Lacetera, N. (2018). Review: Adaptation of animals to heat stress. Animal, 12, 431–444.

Sejian, V., Shashank, C. G., Silpa, M. V., Madhusoodan, A. P., Devaraj, C., & Koenig, S. (2022). Non-Invasive Methods of Quantifying Heat Stress Response in Farm Animals with Special Reference to Dairy Cattle. Atmosphere, 13(10), 1642.

Sejian, V., Silpa, M. V., Devaraj, C., Ramachanndran, N., Thirunavukkarasu, D., Shashank, C. G., Madhusoodan, A. P., Suganthi, R. U., Mylostyvyi, R., Hoffmann, G., Simões, J. C. C., & Bhatta, R. (2024). The welfare of goats in adverse environments. The Welfare of Goats, 273–294.

Ţogoe, D., & Mincă, N. A. (2024). The impact of heat stress on the physiological, productive, and reproductive status of dairy cows. Agriculture, 14(8), 1241.