



## The relationship between blood pH level and changes in lactation performance of heat-stressed Holstein cows

A. F. Washam\*, A. S. Mahdi, H. H. H. Al-Abbasi, A. A. M. Al-Wazeer

*Faculty of Agriculture, University of Kufa, Al Najaf, Iraq*

Article published on October 03, 2024

**Key words:** Heat stressed-Holstein cows, Blood pH, Milk composition, Milk production

### Abstract

The aim of this study was to investigate the correlation between blood pH levels and milk production and composition in Holstein cows during two seasons (winter and summer). Blood and samples were collected from 40 cows in third parity at Taj Al-Nahrain farm station. During summer, the temperature and humidity index (THI) showed that cows exposed to moderate stress with high blood pH levels (7.61-7.96). While in winter, THI values showed that cows below stress levels with low blood pH levels (7.31-7.53). Milk fat, protein and lactose contents and specific density of milk were higher ( $P \leq 0.01$ ) in high blood pH level than low blood pH levels while the non-fat solid content was not affected by blood pH levels. The freezing point of milk increased significantly ( $P \leq 0.01$ ) in cows not exposed to heat stress. Results also revealed that total milk production and persistency were higher ( $P \leq 0.01$ ) with low blood pH than high pH levels. These results showed correlation between blood pH levels and heat stress, it can be used blood pH levels as indicator for heat stress.

\* Corresponding Author: AF. Washam ✉ [alif.altai@uokufa.edu.iq](mailto:alif.altai@uokufa.edu.iq)

## Introduction

Heat stress is defined as an environmental effect that disrupts the balance between heat accumulation and the animal's ability to dissipate heat (Tao *et al.*, 2018). Heat stress represents a problem in the future as greenhouse gases continue to increase in the atmosphere, causing major economic losses, especially in the dairy industry (Collier *et al.*, 2019). Dairy cows are more susceptible to heat stress due to their high metabolic heat production. When heat stress occurs, dairy cows use a variety of physiological and cellular mechanisms to dissipate heat and protect cells from damage. These mechanisms require increased and diverted energy away from other biological processes (Cartwright *et al.*, 2023). The majority of the explanations for the adaptation mechanisms under heat stress have been provided by cutaneous features and respiratory frequency, which it is an important thermoregulation mechanism in birds, goats, sheep and cattle (Das *et al.*, 2016). A higher respiratory rate of 71.5 times/minute was recorded during the summer compared to 38.8 times/minute in the winter in dairy cows (Padilla *et al.*, 2006). It is known that when cows are exposed to heat stress for a long period, the respiratory rate goes through two stages: the stage of rapid, superficial breathing and the stage of slow, deep breathing. The continued rise in temperature leads to the transformation of rapid, superficial breathing into deeper, slower breathing. It has been called the second stage of breathing, as it was observed that cows during the slow, deep breathing phase suffer from a decrease in the amount of carbon dioxide in the blood by 10 mm Hg, accompanied by an increase in blood alkalinity to about 7.7-8. This is due to the decomposition of weak carbonic acid into water and carbon dioxide (Islam *et al.*, 2021). The aim of the current study to determine the relationship between blood pH level and milk production and composition in Holstein cows exposed to heat stress, and the possibility of using the blood pH level as an indicator of the level of thermal stress experienced by the animal, as a tool to predict the animal's productive condition.

## Materials and methods

All animal care and experimental procedures of this study were approved by the bioethics committee at the University of Kufa, Iraq. This study was carried out at Taj Al-Nahrain Farm, a commercial dairy farm located in subtropical area during summer in Qadisiya Governorate, Iraq. Cows were exposed to high temperatures during the day for periods of 10 hours. Forty German Holstein cows of third parity were selected from the herd. During two periods, in winter (January 2023) and summer (July 2023), the level of thermal stresses were determined by measuring the values of the temperature and humidity index (THI) at three times/cow/season according to the method (Segnalini *et al.*, 2011) using the equation  $THI = (1.8 \times \text{ambient temperature} + 32) - (0.55 - 0.55 \times \text{relative humidity}) - [(1.8 \times \text{ambient temperature} + 32) - 58]$ , according to Werama (1990), the levels of thermal stress imposed on cows were determined according to the values of THI. If it is less than 72, the cows do not suffer from heat stress, but they suffer from heat stress if the value of THI ranges from 73 to 79, moderate stress if the value reaches 80 to 89, and severe stress if the THI value reaches greater than or equal to 90. In each period, all cows were fed similar concentrate diet and roughages to meet their requirements. Blood samples were collected from udder vein at two times/cow/season, using a 10 ml medical syringe at different times of the day depending on temperature. The collected samples were transferred into 8-ml gel tubes, cooled in ice water and centrifuged at 14,000 ×g for 15 min to obtain serum. Serum samples were harvested, placed in Eppendorf tubes and stored at -20°C, and transported directly to the laboratory for further analysis. The blood pH was measured using bench type pH meters immediately after collected from udder vein. While milk production data were obtained from the farm's records, where milk production was recorded weekly for the morning and evening milking by the farm. As for the milk components, 10 ml of milk from each cow were collected during two periods / cow / season, and then preserved by refrigeration and transported directly to the laboratory for further analysis.

laboratory analyzes of milk using the Lacto flash (FUNKE GERBER, Germany) which included: Milk protein, fat, lactose, solid non-fat, specific density of milk and freezing point of the milk (FP). Persistency of milk production was determined by dividing the average production of the last three weeks of the period by the average of the first three weeks of the period. All were analyzed statistically using SAS program (SAS, 2012) with completely randomized design, the changes between the

treatments were compared using Duncan's multiple range tests using same program.

**Results and discussion**

During summer, the THI value was 80.46, meaning that they were suffering from moderate stress, and the blood pH level was high (7.61–7.96). While the THI value in winter was 61.45, meaning that the cows did not suffer from any heat stress and the blood pH value low (7.31–7.53).

**Table 1.** Relationship between blood pH level and milk components and characteristics in Holstein cows (Mean ±SEM)

Item	Fat (%)	Protein (%)	Lactose (%)	SNF (%)	FP(°C)	MD (%)
Low blood pH	3.67±0.61 <sup>b</sup>	3.66±0.17 <sup>b</sup>	4.87±0.27 <sup>b</sup>	8.1±0.29	-0.687±24.13 <sup>a</sup>	1.017±0.047 <sup>b</sup>
High blood pH	5.45±0.48 <sup>a</sup>	4.68±0.22 <sup>a</sup>	5.51±0.32 <sup>a</sup>	8.8±0.23	-0.342±23.96 <sup>b</sup>	1.092±0.03 <sup>a</sup>
Significant	**	**	**	NS	**	**

<sup>a,b</sup> letters in the same column with different subscripts are different significantly at P≤0.01; \*\*: P≤0.01; Low blood pH:7.31-7.53 High blood pH:7.61-7.96; SEM: Standard error of means; NS: not significant; SNF: solid non-fat; FP: Freezing point of milk; MD: milk density

**Table 2.** Relationship between blood pH level and milk production characteristics in Holstein cows (Means ±SEM)

Items	Daily milk production (kg/cow)	Total milk production (kg)	Persistency of milk (%)	Output peak (kg)	Season length (d)
Low blood pH	13.59±0.52	509.71±48.41 <sup>a</sup>	2.01±0.670 <sup>a</sup>	16.43±0.55	268.0±11.93
High blood pH	13.89±0.51	391.26±37.60 <sup>b</sup>	1.20±0.065 <sup>b</sup>	16.44±0.54	253.3±11.73
Significant	NS	**	**	NS	NS

<sup>a,b</sup> different letters in same column are significantly different (P≤0.01)

SEM: Standard error of means; NS: Not significant \*\*: (P≤0.01)

Table 1. shows the relationship between blood pH and milk composition and characteristic in Holstein cows. It is clear that cows with high blood pH were significantly higher (P≤0.01) in fat, protein, and lactose contents than cows with low blood pH, while solid non-fat content was not affected by blood pH levels. Milk's freezing point was high significantly (P≤0.01) with high blood pH, while density high (P≤0.01) with low blood pH. Higher specific density of milk in cows exposed to heat stress could be attributed to an increase in milk fat content as a result of decreased milk production and a decrease in water content in the milk. During heat stress, cows increased respiratory rate causes the excretion of the largest amount of CO<sub>2</sub>, which leads to a decrease in the

concentration of carbonic acid in the blood and causes an imbalance of carbonic acid leading to an increase in excretion in urine due to increased drinking of water during heat stress, thus creating an imbalance in the balance necessary to maintain blood pH, causing an increase in blood alkalinity (Conte *et al.*, 2018). The study reveals a correlation between blood pH level and thermal stress in cows, which affects the milk's characteristics and components. During summer the productions of milk decrease as well as its content of water which may lead to an increase in other components and this is somewhat similar to what Ahmad and Al-Khazraji (2021) how found that high THI value in cow exposed to high climate temperature and sunlight.

Cows' total milk production was high ( $P \leq 0.01$ ) with low blood pH than those with high blood pH. While, daily milk yield, peak production, and season length were not alerted by blood pH levels (Table 2). Results revealed that persistency of milk had significantly high ( $P \leq 0.05$ ) with low blood pH. These results are in consistent with the findings of Muschner-Siemens *et al.* (2020) who indicated a decrease in milk production in heat-stressed cows and attributed this to a decrease in the intake and absorption of nutrients and their depletion from the cow's intestines and their spread in the bloodstream to the peripheral tissues that cool the body away from the digestive system. According to Habimana *et al.* (2023) heat stress has a detrimental effect on milk yield because it increases blood flow to the skin's surface, which increases sweat secretion and lowers the volume of blood that travels to the udder. Zhang *et al.* (2016) also found that heat stress alters feeding habits, udder health, and rumen function, all of which lower cattle's milk yield. According to Dahl *et al.* (2016), heat stress causes drop blood glucose concentrations in cows, which lowers the amount of glucose that is supplied to the udder. It also has a detrimental impact on lactose synthesis, which lowers milk production. Furthermore, Fan *et al.* (2019) explained that heat stress reduces the milk synthesis process directly or indirectly by reducing feed intake. In addition, the increased respiratory rates in heat stressed cows, led to decrease  $CO_2$  and increase  $O_2$  in the blood, under heat stress circumstances, metabolic processes in these problems may be altered (Zeng *et al.*, 2023). These results are in harmony with results reported by Bhan *et al.* (2013) who reported that heat stress affected respiration rate of growing Karan Fries cattle during summer over spring.

This can be interpreted as proof that cows during this period have entered the stage of deep breathing, which alters the rate of gas exchange and causes an increase in blood pH. These findings demonstrate the association between blood pH and heat stress placed on the animal, as well as its impact on milk production characteristics, as milk output increased.

## Conclusion

From the result of this study, it can be concluded that heat stress led to increase blood pH and this will lead to change milk components, milk characteristics and reduce total milk production and persistency of milk production due change the blood pH levels, which is considered an indicator to predict the level of milk production and components in cows exposed to heat stress.

## Acknowledgments

We would like to thank the Department of Animal Production - Faculty of Agriculture - University of Kufa and Taj Al-Nahrain Station for their cooperation.

## References

- Ahmad BA, Al-Khazraji WJ. 2021. Relationship of scalp color to production performance and heat tolerance of Holstein cows. *Diyala Agricultural Sciences Journal* **13**(1), 93-99.  
<https://doi.org/10.52951/dasj.21130109>.
- Bhan C, Singh SV, Hooda OK, Upadhyay RC, Beenam B. 2013. Influence of temperature variability on physiological, hematological and biochemical profiles of growing and adult Karan Fries cattle. *The Indian Journal of Animal Sciences* **83**(10), 1090-1096.
- Cartwright SL, Schmied J, Karrow N, Mallard BA. 2023. Impact of heat stress on dairy cattle and selection strategies for thermotolerance: A review. *Frontiers in Veterinary Science* **10**, 1198697.
- Collier RJ, Baumgard LH, Zimelman RB, Xiao Y. 2019. Heat stress: physiology of acclimation and adaptation. *Animal Frontiers* **9**(1), 12-19.
- Conte G, Ciampolini R, Cassandro M, Lasagna E, Calamari L, Bernabucci U, Abeni F. 2018. Feeding and nutrition management of heat-stressed dairy ruminants. *Italian Journal of Animal Science* **17**(3), 604-620.  
<https://doi.org/10.1080/1828051X.2017.1404944>.

- Dahl GE, Tao S, Monteiro APA.** 2016. Effects of late-gestation heat stress on immunity and performance of calves. *Journal of Dairy Science* **99**(4), 3193-3198.  
<https://doi.org/10.3168/jds.2015-9990>.
- Das R, Sailo L, Verma N, Bharti P, Saikia J, Kumar R.** 2016. Impact of heat stress on health and performance of dairy animals: A review. *Veterinary World* **9**(3), 260-268.  
<https://doi.org/10.14202/vetworld.2016.260-268>.
- Fan C, Di S, He T, Hu R, Lei R, Ying Y, Su Y, Cheng JB.** 2019. Milk production and composition and metabolic alterations in the mammary gland of heat-stressed lactating dairy cows. *Journal of Integrative Agriculture* **18**(12), 2844-2853.  
[https://doi.org/10.1016/S2095-3119\(19\)62834-0](https://doi.org/10.1016/S2095-3119(19)62834-0).
- Habimana V, Nguluma AS, Nziku ZC, Ekine-Dzivenu CC, Morota G, Mrode R, Chenyambuga SW.** 2023. Heat stress effects on milk yield traits and metabolites and mitigation strategies for dairy cattle breeds reared in tropical and sub-tropical countries. *Frontiers in Veterinary Science* **10**, 1121499.  
<https://doi.org/10.3389/fvets.2023.1121499>.
- Islam MA, Lomax S, Doughty AK, Islam MR, Thomson PC, Clark CE.** 2021. Revealing the diversity in cattle behavioural response to high environmental heat using accelerometer-based ear tag sensors. *Computers and Electronics in Agriculture* **191**, 106511.  
<https://doi.org/10.1016/j.compag.2021.106511>.
- Muschner-Siemens T, Hoffmann G, Ammon C, Amon T.** 2020. Daily rumination time of lactating dairy cows under heat stress conditions. *Journal of Thermal Biology* **88**, 102484.  
<https://doi.org/10.1016/j.jtherbio.2019.102484>.
- Padilla L, Matsui T, Kamiya Y, Kamiya M, Tanaka M, Yano H.** 2006. Heat stress decreases plasma vitamin C concentration in lactating cows. *Livestock Science* **101**(1-3), 300-304.  
<https://doi.org/10.1016/j.livprodsci.2005.12.002>.
- SAS.** 2012. Statistical Analysis System, Users Guide. Statistical. Version 9th ed. SAS. Inst. Inc., Cary, NC, USA.
- Segnalini M, Nardone A, Bernabucci U, Vital A, Ronchi B, Lacetera N.** 2011. Dynamics of the temperature-humidity index in the Mediterranean basin. *International Journal of Biometeorology* **55**(2), 253-263.
- Tao S, Orellana RM, Weng X, Marins TN, Dahl GE, Bernard JK.** 2018. Symposium review: The influences of heat stress on bovine mammary gland function. *Journal of Dairy Science* **101**(6), 5642-5654.
- Wierama F.** 1990. The Temperature-Humidity Index (THI) Matrix–Critical Temperature Zones for Dairy Cattle. Department of Agricultural Engineering, The University of Arizona, Tucson, Arizona.
- Zeng J, Cai J, Wang D, Liu H, Sun H, Liu J.** 2023. Heat stress affects dairy cow health status through blood oxygen availability. *Journal of Animal Science and Biotechnology* **14**(1), 112.  
<https://doi.org/10.1186/s40104-023-00915-3>.
- Zhang Y, Wang QC, Yu H, Zhu J, de Lange K, Yin Y, Wang Q, Gong J.** 2016. Evaluation of alginate–whey protein microcapsules for intestinal delivery of lipophilic compounds in pigs. *Journal of the Science of Food and Agriculture* **96**(8), 2674-2681.  
<https://doi.org/10.1002/jsfa.7385>.