Effects of Heat Stress Induced Physiological Changes on the Productivity of Dairy Cattle

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Thesis submitted to the faculty of the Virginia Polytechnic Institute and State University

in partial fulfillment of the requirements for the degree of

Master of Science

In

Animal and Poultry Sciences

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May 7th, 2024

Blacksburg, Virginia

Keywords: Heat Stress, Holsteins, Blood glucose concentrations

Relationships between circulating glucose concentrations and productivity in heat-stressed dairy cattle

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ABSTRACT

Dairy cattle exposed to summer heat stress (HS) typically exhibit altered metabolic characteristics, including hyperinsulinemia concurrent with hypoglycemia. The reasons for this change in glycemic status and its consequences are currently unknown. This project aimed to examine the relationship between summer HS, blood glucose concentrations, and milk production in primiparous (n=26) and multiparous (n=27) Holstein cows. Glucose was measured in coccygeal vein blood twice weekly using a hand-held glucometer (Abbott Precision Xtra) from May through July. Rectal temperatures were also collected. The days surrounding sample collections were categorized as having no HS (0 h above 68 Thermal Humidity Index [THI]), moderate HS (>0 to 8 h above 68 THI) or severe HS (>8 h above 68 THI). A subset of cows with ≥5 blood glucose measurements spanning different THI categories were categorized as hypoglycemic (n=8 primiparous; n=9 multiparous) or non-hypoglycemic (n=6 primiparous; n=5 multiparous) based upon blood glucose concentrations on no-HS days and severe-HS days (hypoglycemic if sustained reduction on severe-HS days). When analyzed together, milk production did not differ between those categorized as hypoglycemic and non-hypoglycemic. When separated based upon parity, there was a tendency for primiparous hypoglycemic cows to produce more milk than their non-hypoglycemic counterparts ($P<0.15$). Variability in milk production measured as standard deviation across no-, moderate- and severe-HS days did not differ based upon glycemic status but was greater in multiparous than primiparous cows ($P<0.01$). Rectal temperatures were greater on severe-HS days than on no- or moderate-HS days ($P<0.01$) and were positively correlated with the number of h of HS in a day ($r=0.45$; $P<0.01$). Neither milk nor glucose differed based upon the category of HS severity. Interestingly, however, glucose was negatively correlated with milk production on the day of measurement ($r=-0.24$; $P<0.01$) and day after measurement ($r=-0.23$; $P<0.01$). These results are an initial indication that hypoglycemia during HS is related to milk production, particularly in primiparous cows.

Relationships between circulating glucose concentrations and productivity in heat-stressed dairy cattle

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GENERAL AUDIENCE ABSTRACT

Dairy producers lose production and profitability when their dairy cows experience heat stress. Heat stress in dairy cattle during the summer months causes declines in milk production and reproductive performance, which may be at least partially due to the unique metabolic changes observed in heat-stressed dairy cattle. One of these changes is a reduction in blood glucose that has been linked with productivity in previous studies. Measuring blood glucose concentrations may be a useful tool for measuring individual cow responses to heat stress, and ultimately lead to a better understanding of these changes. The work described here aims to explore the relationship between both summer heat stress and blood glucose concentrations in primiparous (calving =1) and multiparous (calving >1) Holstein cows. A total of 53 cows were used, 26 being primiparous and 27 being multiparous. Rectal temperatures, blood glucose measurements and milk production records were collected. A subset of cows were categorized as either hypoglycemic or non-hypoglycemic based on glucose concentrations on non-heat stress and heat stress days. When all cows were analyzed together, milk production did not differ based on glycemic category. When separated based upon parity, there was a tendency for cows that had calved once and were hypoglycemic to produce more milk than their non-hypoglycemic contemporaries. Rectal temperatures were greater on

severe-heat stress days than on no- or moderate-heat stress days and were positively correlated with the number of hours of heat stress in a day. Neither the quantity of milk produced, nor blood glucose differed based upon the severity of heat stress. Interestingly, however, blood glucose was negatively correlated with the quantity of milk produced on the day of blood collection and day after blood collection. These Results indicate that measuring blood glucose concentrations could be a useful tool in predicting individual cow’s responses to heat and their ability to sustain productivity in summer months.

DEDICATION

I would like to dedicate this thesis to my mother and grandmother for always believing in me and my success in my academic career. They are true prayer warriors always in my corner to extend wisdom, love, and care when I need it. These ladies are truly my inspiration.

ACKNOWLEDGEMENTS

I would like to send a special thank you to Dr. Michelle Rhoads for allowing me to pursue new knowledge on animal welfare and more insight into the industry. I am forever grateful to the skills that I learned through my time as a graduate student. I would like to thank my committee for always being supportive and giving me guidance when needed- Dr. Alan Ealy and Dr. Jamie Stewart. I would like to thank my lab mate Kamryn Joyce as well as those who helped along in the project, Sarah Newman, and Nikki Tabatabai for making this time an unforgettable experience. I would also like to express gratitude to the grad students – Jessica Keene and Endya McKinley for always being a helping hand and making my graduate journey that much more enjoyable. The biggest thank you I would like to give is to my assistantship program George Washington Carver Program, for not only helping with financial aspect of school but also for the knowledge and lifelong friendships that I have made through this program. And finally, my friends and family for the unimaginable support and love that has been giving unto me since the beginning.

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List of Abbreviations

FSH: Follicle Stimulating Hormone

GLUT: Glucose Transporters

LH: Luteinizing Hormone

MP: Multiparous

NEB: Negative Energy Balance

NEFA: Non-Esterified Fatty Acid

PP: Primiparous

THI: Temperature Humidity Index

VFA: Volatile Fatty Acids

Chapter 1: Literature Review

Introduction

Environmental impacts, such as global warming, are increasingly affecting consumers and producers daily. These impacts, in turn, have significant implications for food animal production, with dairy cattle being particularly vulnerable. Dairy cattle play a vital role in food production for the growing global human population. However, the rising temperatures, especially during summer months, pose serious challenges for milk production and reproductive efficiency of dairy cattle.

While there are decades of research on heat mitigation strategies, one approach currently under investigation for monitoring heat stress in dairy cattle involves blood glucose monitoring. While a calculated measurement of weather called the temperature humidity index (THI) serves as an indicator of heat stress at the environmental level, glucose concentrations could be an indicator of heat stress at the cow level. This measurement of weather conditions called temperature humidity index is not only a method for detection of metabolic imbalances but could also be associated with immune function and reproductive performance. Continued research in this area is a primary focus of the work described in this thesis and holds promise for utilizing glucose monitoring as a tool for precision management of heat-stressed dairy cows.

This literature review provides an overview of the importance of addressing the detrimental effects of heat stress on dairy cattle. Specifically, it examines the known effects of heat stress, with an emphasis on changes in blood glucose and the related implications.

Climate Change and Agricultural Animal Production

Environmental changes exert a significant impact on agricultural productivity and related commodities. Climate change is an ongoing issue that the worldwide population has grappled with for several decades. It is defined as the "long-term alteration to the typical weather patterns that have come to characterize local, regional, and global climates on Earth" (*What is Climate Change*, n.d). Human activities, particularly the burning of fossil fuels, contribute significantly to climate change by escalating the presence of heat-trapping greenhouse gases in the atmosphere (*What is Climate Change*, n.d). Factors such as prolonged seasons and heavier rainfall contribute to runoff and flooding, potentially leading to nutrient loss. The USDA suggests that heavier rainfall could negatively impact nutrient retention by up to 40% by 2050 and 87% by 2100, thereby increasing the risks of soil erosion, compaction, and nutrient depletion (USDA, n.d). Such events can significantly impact livestock productivity, particularly dairy cattle, thereby affecting global food security (Sungkhapreecha et al., 2022).

Heat Stress

Heat stress can be defined in multiple ways. The word stress, in relation to the environment, encompasses both the conditions surrounding the animal and the animal's ability to adapt to the environment (Collier & Collier, 2012). It is also defined as any condition that can alter the resting state of a system, whether physiologically or biologically, potentially prompting an adaptive response (Collier & Collier, 2012). When agricultural animals are exposed to high temperature conditions, they may experience various physical symptoms, including but not limited to reduced milk production, pregnancy loss, and economic losses related to production and animal welfare (Collier & Collier, 2012).

Environmental conditions affect the dissipation of body heat in lactating cows. The ability of an organism to regulate its body temperature within predetermined limits, even in the face of considerable ambient temperature variations, is known as thermoregulation (Collier & Collier, 2012). The thermal neutral zone of dairy cattle ranges from 16° C to 25° C (33° F to 77° F), with cows maintaining a body temperature of 38.4° C to 39.1° C (Das et al., 2016). Traditionally, air temperatures exceed this range (Das et al., 2016). Evaporation serves as one of the primary mechanisms for the animals to reduce internal body temperature when ambient temperatures surpass their body temperature. Thermoregulation is essential for lactating cows to maintain homeostasis and homeothermy by effectively managing body heat accumulation and dissipation. Heat stress represents a critical environmental challenge, underscoring the significance of understanding the physiological responses to seasonal stressors that directly and indirectly affect animal well-being and productivity.

Humidity and the Temperature Humidity Index

Ambient temperature by itself is often not a sufficient indicator of the risk that the environmental conditions will (or will not) cause stress in an animal. Assessing relative humidity alongside temperature is often a more accurate approach for evaluating environmental conditions as they relate to thermal stress in animals. Therefore, THI is calculated from both temperature and relative humidity and used to predict when the animal is at risk for developing heat stress (Dikmen & Hansen, 2009).

The THI is used as an indicator to predict if exposed animals are able to dissipate heat regularly. When the THI exceeds a certain threshold, animals are at risk for hyperthermia which, if not managed properly, could lead to death (Herbut et al., 2018).

According to previous studies, an average cow remains unaffected by heat stress when the THI is below 72, experiences mild heat stress between 72 and 79, moderate heat stress between 80 and 89, and severe heat stress at 90 or above (Liu et al., 2019; Armstrong, 1994). Despite these widely used thresholds another study found that milk production decline can occur as low as a THI of 68. Therefore, further studies are needed to better assess THI cut offs for heat stress by evaluating known physiological outcomes (Zimbelman et al., 2009).

Breed Differences

When discussing heat stress in dairy cattle, it's important to consider breed differences. In the United States, Holsteins and Jersey cows are the most prevalent dairy cattle breeds, each valued for distinct reasons. Holsteins are favored for their high milk production capacity, while Jerseys typically yield lower quantities of milk with superior characteristics such as higher fat, protein, vitamins, and minerals (Lim et al., 2021). Jersey cows are smaller than Holsteins and produce less milk overall (Lim et al., 2021). Holsteins are typically more resistant to colder temperatures but are more susceptible to heat stress.

Bos taurus and Bos indicus cattle also differ in heat tolerance. Bos indicus breeds are known for their heat resistance, having less severe reductions in feed intake, growth rate, milk yield and reproduction function, while Bos taurus breeds are less heat tolerant (Hansen, 2004). This could be because Bos taurus breeds originate from temperature climates such as eastern Europe and are more tolerant to cooler temperatures. Bos taurus breeds are more known for the meat quality than milk production, which is beneficial to southern states like Texas, bit considering that these breeds are susceptible to heat, Bos indicus breeds are the preferred breeds to use in milk production due to their tolerance to heat especially in southern states. Individual factors such as age, body composition, breed, and environmental conditions like wind, shade, and water availability all influence the response to heat stress (Capela et al., 2022).

Cattle in countries with moderate climates, such as those in the European Union, are also vulnerable to heat stress. For example, in the southwest regions of the United Kingdom, where Bos taurus breeds such as Holstein-Fresinas are prominent, dairy farms experience an annual production loss of 2.4% due to heat stress (Habimana et al., 2023). In tropical or subtropical areas, Bos indicus breeds are preferred for their genetic heat tolerance and resilience, influencing housing decisions and driving future advancements in cost-effective solutions for dairy production in hot and humid climates (Habimana et al., 2023).

Parity

Primiparous cows (PP), which have undergone only one calving, seem to exhibit differences in their response to heat stress than multiparous cows (MP) that have experienced multiple calvings. Parity affects many variables such as milk yield, non-heat related stress, and welfare (Davis et al., 2023). A study conducted by Davis (2023) investigated parity and its effects on behavior, production, in non-heat related stress conditions. In this study, PP cows when placed in commitment pens and in the milking parlor, showed signs of increased stressed levels as well as increased heart rate variability compared to their MP counterparts (Davis et al., 2023). The MP cows tended to increase stepping behavior in entering the milking parlor, which is an indicator of minor stress and increased lying times. Overall, this study indicated that PP cows were more stressed than their MP counterparts, which could be related to social competition (Davis et al., 2023). While parity research itself is limited, studies like the one mentioned give insight into how parity is related to production of the animal as well as animal welfare.

Dairy cows, in general, experience a negative energy balance (NEB) during early lactation due to reduced feed intake and increased metabolic demands (Wathes et al., 2007). While research on parity-specific differences in heat-stressed dairy cattle is limited, some studies suggest that PP cows are more susceptible to metabolic changes when compared to MP cows. This susceptibility to metabolic changes during heat stress could be related to the fact that cows generally calve for the first time at about 24 months of age in order to maximize profitability (Wathes et al., 2007). Considering cows are still growing at this age, their metabolic demands at the beginning of lactation will differ compared to MP cows, primarily due to greater nutrient requirements associated with continued growth during lactation (Coffey et al., 2006; Wathes et al., 2007).

Performance Measurement in Cattle

Diet plays an important role in addressing heat stress in dairy cattle. Feed is particularly vital as it directly impacts milk production and accounts for approximately 53% of the total cost of production (Manthey et al., 2016). Cows experiencing heat stress reduce their feed intake which contributes to the observed reduction milk production during heat stress. Interestingly, however, decreased feed intake during periods of heat stress is not the primary factor affecting milk yield. A study by Rhoads and others (2009), examined this relationship by comparing heat stressed cows to pair-fed counterparts. Cows that were heat stressed reduced feed intake by 37% and pair-fed cow intake was limited accordingly while they remained in thermoneutral conditions (Rhoads et al., 2009). Considering that all cows were consuming the same amount of feed, it was expected that milk production would be similar between the two treatment groups. Instead, however, heat-stressed cows produced considerably less milk than the pair fed cows (Rhoads et al., 2009). The researchers determined that while feed intake affects milk production during heat stress, it only accounts for approximately 35% of the reduction in milk yield.

The time of day during feeding can also greatly affect animal performance during summer months. While evening feeding practices are not extensively studied, authors of one study found that cows fed during the night experienced a decline in energy expenditure and increased energy efficiency for milk production versus those during the day (Aharoni et al., 2005). In another study, Calamari (2013) found that cows fed in the morning were more susceptible to heat stress than those who were fed in the evening. While morning-fed cows had increased rectal temperatures and respiratory rates over evening-fed cows, milk yield was not affected (Calamari et al., 2013).

Impact of Heat Stress on Milk Production

The dairy industry contributes to two-thirds of the United States milk supply and one-third of the world's milk production, generating revenue of $35.5 billion of revenue in 2012 (Gunn et al., 2019). However, due to heat stress, the dairy industry has experienced economic loss of between $897 and 1500 million annually and a total annual loss of $1.5 billion (St. Pierre et al., 2003). This economic pressure necessitates proactive measures to mitigate heat stress and maintain optimal body temperature in cattle to safeguard against potential milk loss.

Milk production during heat stress is influenced by various factors, including parity, lactation stage, and metabolic heat production (Tao et., 2020). Research suggests that heat stress exerts the most significant impact during the mid-stage of lactation, with the expectation that PP cows experiencing heat stress will have smaller milk yield loss compared to their MP counterparts (Tao et al., 2020). Moreover, as homeotherms, dairy cows struggle to maintain body temperature while producing high milk quantities under conditions of elevated metabolic heat exposure (Tao et al., 2020).

Reproductive Factors Affected by Heat Stress

Heat stress profoundly affects reproductive activity, posing concerns for dairy producers worldwide. The detrimental impact extends across all aspects of fertility and fetal development, potentially resulting in reproductive loss. Heat stress can affect the growth of the oocyte, follicular development, estrus duration, uterine function, conception, gestation, and other aspects of reproductive performance (Jordan, 2003; Bernabucci et al., 2010). Fertility is an important factor in sustainable livestock production, and heat stress is a prevalent issue in the decline of fertility within both beef and dairy industries. Conception rates decline from 40 to 60 percent in the cooler months to roughly 10 to 20 percent or lower in the summer months, depending on the severity of heat stress (Wolfenson et al., 2000). This characteristic decline in conception rates is the result of detrimental effects on multiple reproductive tissues and processes.

Heat stress also affects the secretion of gonadotrophin-releasing hormone, leading to alterations in the levels of luteinizing hormone (LH) and follicle-stimulating hormone (FSH) (Miękiewska et al., 2022). The relationship between heat stress and LH secretion has generated conflicting findings in the literature. Some studies have reported unchanged LH concentrations during heat stress, while most studies have reported decreased LH secretion (Khan et al., 2023; Howell et al., 1994). These discrepancies may arise from variations in heat stress conditions and other methodological differences. Likewise, there is a lack of consensus regarding FSH secretion during heat stress. Many conclude that FSH secretion is greater during heat stress because of decreased inhibin production from heat comprised follicles (Renis & Scaramuzzi, 2003).

Heat stress is known to disrupt oocyte physiology, which can negatively impact fertility (Miękiewska et al., 2022). The negative effects of elevated temperatures on the developing oocyte include depleted maturation and altered molecular and cellular components, which critically impair its function. A study by Edwards et al. (2005), investigated the *in vivo* effects of heat stress on bovine oocytes. In this experiment, ovaries purchased a commercial abattoir, and the collected cumulus-oocyte-complexes were randomly allocated to treatment groups, control and heat stressed. Heat-stressed oocytes exhibited accelerated in vitro maturation compared to the control group, but when compared to nuclear maturation or early insemination, the heat stressed oocytes and the controlled oocytes did not differ (Edwards et al., 2005).

The detrimental effects of heat stress on follicular development ultimately compromise reproductive performance in cattle. Though follicle development can be affected at any stage, the primary stages of follicular development are especially vulnerable to heat stress (De Rensis et al., 2021). The primary stage is when the zona pellucida material is distributed around the oocyte and the cortical granules are formed, after which follicles either enter maturation or undergo atresia (Fair, 2003). In early antral development, heat stress will cause the size of the dominant follicle present to be reduced, which attenuates dominance and increases the number of large sized non-dominant follicles (De Rensis et al., 2021). The reduced size of the dominant follicle due to elevated THI and heat stress will lead to a smaller final size of the pre-ovulatory follicle (Jitjumnong et al., 2020). This reduction in follicle size is related to decreased estradiol and progesterone production from the follicular granulosa cells (Miękiewska et al., 2022).

The smaller size of the ovulatory follicle means that there are fewer cells available to form the corpus luteum after ovulation. As a result, the corpus luteum produces lower levels of progesterone during heat stress, which can cause abnormalities in embryonic development and implantation failure (Gupta et al., 2022). Heat stress adversely affects early embryonic development following fertilization, as quickly as day 1 of pregnancy (Ealy et al., 1993). During the early cleavage stage of especially susceptible to elevated temperatures (Satrapa et al., 2013). During this time, a heat shock protein, traditionally heat shock protein 70, is released to protect the cells and combat the stressor, which is heat stress (Miękiewska et al., 2022). Heat stress also affects the blastocyst phase after the zygotes are exposed, especially when the mother's rectal temperature is above 1 to 2 degrees of normal temperature (Miękiewska et al., 2022).

Gestation, or pregnancy, is another reproductive phase affected by heat stress. Heat stress during late gestation affects calf development and health (Dahl et al., 2016). Early gestation is the period in which embryonic development occurs. Heat stress during early gestation increases the rates of early embryonic death and pregnancy loss (Kasimanickam & Kasimanickam, 2021). Late gestation encompasses the period where most of the calf growth will occur, as at the end of the first 7 months of gestation, the calf is approximately 40% of its birth weight (Bauman & Bruce Currie, 1980). In late gestation, maternal heat stress can compromise fetal muscle growth, fat deposition, and aspects of maternal lactation, thereby affecting calf health, immunity and development (Tang et al., 2022). On the maternal side, colostrum is an important supplement for the neonatal calf to ensure adequate immunity. Research conducted by Nardone, and co-workers (1997) determined that heat-stressed cows had lower IgG and IgA concentrations in their colostrum, as well as lower mean percentages of total protein, casein, lactalbumin, fat and lactose (Nardone, 1997).

Overall, the effects of heat stress on reproductive function of dairy cattle are detrimental for the cows, producers and consumers due to negative impacts on productivity and dairy food production. All aspects of reproduction are affected by heat stress in one way or another. The reproductive performance of heat-stressed dairy cows should be closely monitored and managed to optimize productivity in the warm seasons of the year.

Glucose

Overview

In ruminating species, glucose is primarily derived through the process of gluconeogenesis, mainly occurring in the liver (Lohrenz et al., 2010). Glucose serves as the primary precursor for lactose synthesis in dairy cattle, and to sufficiently synthesize significant amounts of milk, glucose must be produced in consistent and abundant quantities (Abbas et al., 2020). The overall quantity of milk produced is directly related to lactose as its osmoregulatory characteristics are responsible for water uptake in the mammary gland (Abbas et al., 2020). Glucose absorption by mammary tissue necessitates specific carrier proteins due to its hydrophilic nature, preventing it from freely traversing the lipid bilayer (Abbas et al., 2020). The demand for glucose extends beyond lactation, as it is vital for various organs and tissues throughout the body, especially mammary tissue (Abbas et al., 2020).

Circulating insulin concentrations are generally related to the availability of glucose within the body. When glucose increases, insulin is then synthesized and secreted to help facilitate glucose uptake into tissues (Herbein et al., 1985). During periods of heat stress however, this relationship is somewhat altered in lactating dairy cattle. Reduced feed intake results in relatively low glucose concentrations, which would normally cause circulating insulin concentrations to also be low. Instead, however, circulating insulin concentrations increase in comparison to pair-fed counterparts (Wheelock et al., 2010). The change in insulin is counterintuitive since insulin concentrations are generally related to glucose concentration in the blood (Stewart et al., 2022).

During periods of elevated heat, animals undergo molecular, physiological, and biochemical adaptations to cope with prolonged exposure (Abbas et al., 2020). As a result of these alterations, heat stress negatively impacts circulating glucose concentrations and its availability for lactose and milk synthesis. Dairy cattle have been extensively studied regarding the effects of heat stress on blood glucose levels. For dairy cattle and other ruminant animals, glucose originates from dietary intake and hepatic gluconeogenesis (Abbas et al., 2020). The rumen contains microorganisms that are able to digest fibrous feed, which ultimately allows them to partially digest plant cellulose and hemicellulose, leading to the formation of volatile fatty acids (VFA) like propionate, butyrate and acetate (Abbas et al., 2020). Carbohydrates are important for gluconeogenesis as they are fermented into VFA in the rumen (Young, 1977). The VFA, specifically propionate, are used for glucose synthesis in the liver. Hepatic gluconeogenesis represents a substantial portion (75-90%) of glucose needs for downstream physiological functions (Abbas et al., 2020).

The relationship between heat stress and blood glucose levels in dairy cattle is affected by reduced feed intake and energy balance, leading to destabilization and subsequent modulation of energetic metabolism (Wheelock et al., 2010). Reduced feed intake by heat stressed dairy cattle contributes to decreased milk synthesis, but only accounts for approximately 35% of the reduction (Wheelock et al., 2010). Heat-stressed cows experience low milk and often exhibit NEB due to reduced feed intake (Sammad et al., 2020). During thermoneutrality, early lactating cows undergo NEB to support increased demand in milk supply, in which low glucose levels and high non-esterified fatty acids (NEFA) are traditionally seen, at elevated levels (Sammad et al., 2020). The NEB reduces insulin concentrations, which results in a lipolytic effect that then stimulates the export of NEFA from the adipose tissue while inhibiting insulin activity (Sammad et al., 2020).

The downstream impacts of heat stress on gluconeogenesis ultimately affect overall metabolic health in cows. Traditional indicators like THI indicate the potential for thermal heat stress but are not completely predictive of the typical metabolic changes observed in heat stressed-dairy cattle (Yue et al., 2020). Heat-stressed cows exhibit high insulin activity, immobilization of adipose tissue, and alteration of glucose-sparing mechanisms (Sammad et al., 2020). These metabolic alterations not only exacerbate stress but also diminish milk yield during heat stress episodes (Sammad et al., 2020).

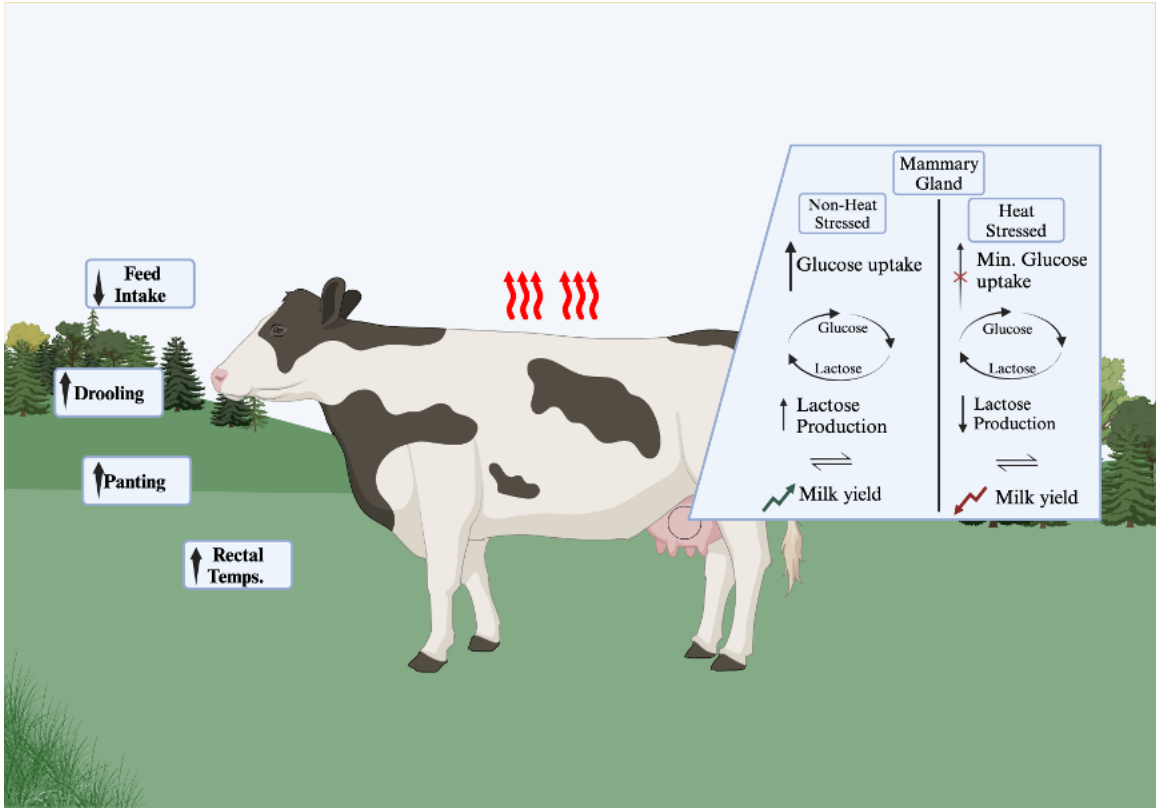

Figure (1): Diagram of dairy cattle experiencing heat stress and glucose uptake through the mammary gland. Side-by-side reference of normal glucose uptake would convert to vs. what is thought to happen to the cow while undergoing heat stress. Created with BioRender.com

Secondary Health Impacts: Mastitis

Dairy cattle experiencing heat stress are susceptible to developing secondary health issues such as respiratory infections, mastitis, ketosis, and more. Mastitis is an infectious disease of the mammary tissue that causes pain and swelling and decreases milk expulsion. It is common in the dairy industry and causes significant financial losses attributed to reduced production yield and milk quality. The substantial economic losses are estimated at over 40 billion USD annually in the dairy industry with an estimated loss of $147 per cow (Neculai-Valeanu & Ariton, 2022; Cheng & Han, 2020).

Mastitis can manifest in three forms: clinical, subclinical, and chronic. Clinical mastitis presents noticeable abnormalities such as redness, swelling, and fever in dairy cows, while subclinical mastitis, though less visible, poses significant financial risks if left untreated. Chronic mastitis involves prolonged inflammatory responses at irregular intervals (Cheng & Han, 2020). Somatic cell count is an indicator for both infection and clinical mastitis (Morse et al., 1988), and increased THI values are related to increased somatic cell counts, indicating signs of infection (Vitali et al., 2020). Clinical mastitis is more prevalent during heat stress due to higher temperatures creating an environment for udder infection (Morse et al., 1988). Overall, mastitis poses a significant health concern for dairy farm operations and is exacerbated during periods of heat stress.

Secondary Health Impact: Ketosis and Fatty Liver Disease

Ketosis, characterized by elevated circulating ketone bodies in early lactation, occurs due to increased milk production coupled with reduced dry matter intake (Duffield, 2020). Ketones serve as crucial energy sources for peripheral tissues in ruminants when carbohydrate consumption declines. During heat stress, ketosis is exacerbated due to the decline in feed intake and carbohydrates consumption, which results in low glucose production (Guliński, 2021). Major ketone bodies include acetone, acetoacetate, and beta-hydroxybutyrate, with the latter also contributing to milk fat synthesis (Duffield, 2020). Subclinical ketosis, a common form, involves excessive ketone circulation without overt clinical signs. Physical manifestations of ketosis include decreased appetite, dry feces, reduced milk yield, and weight loss, alongside other signs like excessive licking and blindness (Duffield, 2020).

Fatty liver disease, a metabolic disorder, can also coincide with ketosis. If not managed properly, ketosis can turn into fatty liver disease. This disease occurs when liver lipid uptake surpasses lipid use (Bobe et al., 2004). Excess lipids are stored as triacylglycerol in the liver, impairing its metabolic functions, and are commonly observed in recently calved cows (Bobe et al., 2004). This condition often correlates with reduced feed intake, impacting the availability of nutrients needed for lactation requirements and leading to the accumulation of NEFA from adipose tissue, exacerbating NEB (Bobe et al., 2004). Cows with moderate to severe fatty liver disease experience heightened NEB particularly when heat stress reduces feed intake (Bobe et al., 2004).

Although fatty liver disease may be caused or worsened by the effects of heat stress, affected cows typically perform comparably to unaffected counterparts in terms of milk production (Bobe et al., 2004). While this disease can be diagnosed with a liver biopsy or biochemistry panel, this would not be a practical option for producers on farms for routine monitoring. Nevertheless, considering its association with feed intake, it is a malady that is likely more prevalent during heat stress. Nevertheless, considering its association with feed intake, it is a malady that is likely more prevalent during heat stress.

Secondary Health Impact: Respiratory Disease

Respiratory diseases are a significant concern for lactating dairy cattle and are exacerbated by prolonged heat exposure. Elevated temperatures, humidity, and solar radiation increase body temperature, prompting physiological adaptations like increased peripheral circulation vasodilation and sweating to dissipate heat (Idris et al., 2021). Sweating and panting serve as primary cooling mechanisms for cattle, offering observable signs of overheating. Respiratory rates, reaching up to 200 breaths per minute in heat-stressed cows, indicate an imbalance between heat accumulation and dissipation, and can be used as an indicator for early detection of heat stress (Idris et al., 2021).

Secondary Health Impact: Immunological Response

Heat stress significantly impacts the immune system in dairy cattle (Gupta et al., 2022), with a THI exceeding 68 considered stressful (Brown et al., 2016). Heat stress affects adaptive immune systems factors such as cell mediated immune responses and humoral responses. Regarding cell mediated responses, heat stress affects the peripheral blood mononuclear cells under in vitro conditions, however the physiological state of the animal depends on factor regarding its effects (Bagath et al., 2019). The humoral immune response is also dampened by heat stress, causing cattle to be more susceptible to infection (Bagath et al., 2019). The immune system consists of innate and adaptive responses, with innate immune system functioning independent of exposure to pathogens, and the adaptive immune system dependent of exposure to pathogens (Gupta et., 2022). Less is known about the ways heat stress affects the immune systems of growing heifers, but it is assumed that heat tolerance and heat dissipation decrease with age (Gupta et al., 2022).

Immunological evaluations are crucial for assessing heat stress impacts on dairy cattle, as immune function influences their ability to combat infections like mastitis, respiratory diseases and more. Addressing heat stress through improved management practices is essential for maintaining animal well-being amid rising temperatures. Producers and veterinarians should prioritize measures to mitigate heat stress and support the overall health of dairy cattle in increasingly challenging environmental conditions.

Management and Effective Practices

Management Practices

Farmers and producers employ diverse management practices tailored to their farms, production yields, and geographic locations. It's important to acknowledge that what works for one farm may not be applicable to another, even though all have an end goal of prioritizing animal welfare while maintaining productivity. Geographically distinct regions face unique challenges in dairy cow production. In the southeast, characterized by hot humid summer conditions, researchers have forecasted significant milk production losses due to high seasonal temperatures, humidity, and rainfall (West, 2003). The northeastern United States' agricultural economy greatly depends on dairy production, with milk sales surpassing $7.5 billion in 2014 alone (Hristov et al., 2018). Sales of dairy products account for 32% of all farm income in the region (Hristov et., 2018). In the coming years, rising temperatures and increased humidity will amplify challenges related to heat stress in this important dairy producing region, highlighting the need for effective mitigation strategies (Hristov et al., 2018).

The dairy industry faces challenges in maintaining longevity and meeting the growing demand for livestock products. Projections indicate that by 2050, livestock products, including dairy, will need to increase by 70 percent on a global scale (Cartwright et al., 2023). Achieving this goal will be challenging for many reasons, necessitating strategies to address challenges such as heat stress, which disproportionately affects the dairy industry compared to other livestock sectors (Cartwright et al., 2023). Research on mitigation strategies is crucial for sustaining dairy production amid rising temperatures and ensuring future expansion to meet the needs of a growing global population.

Heat Abatement Systems

Heat abatement systems are essential components of the dairy industry, profoundly impacting cows' comfort and productivity. Over the past two decades, significant technological and constructive advancements have revolutionized dairy farming facilities, from animal housing to feeding and milking processes, innovations have continually improved. However, alongside these advancements, pressing issues have emerged, with global warming being particularly prevalent.

Common methods to combat heat stress can be categorized into two approaches: environmental modification and enhancing heat exchange between cows and their environment (Fournel et al., 2017). Effective housing is pivotal for ensuring the well-being and health of dairy cattle. Producers employ various strategies and tools, such as fans and ventilation systems, to mitigate heat stress during warmer months. Ventilation systems provide a cool and clean environment conducive to animal welfare, with options for natural and ambient ventilation conditions (Mondaca & Cook, 2019). However, factors like wind shadows created by nearby buildings, land availability, and barn orientation influence the functionality of mechanical ventilation, making it necessary for producers to make informed choices based on their specific circumstances (Mondaca & Cook, 2019).

Producers must tailor their strategies based on factors such as farm size, location, and housing methods. Mechanical interventions like misters, sprinklers or fans are commonly used to mitigate environmental conditions or improve heat exchange, depending on the specific needs of each farm (Fournel et al., 2017). Research has shown that, generally, cows that are shaded do experience physiological impacts of heat stress but not on the same level as non-shaded. In one study shaded cows experience lower rectal temperatures (38.7 °C vs. 39.3 °C, respectively) (Fournel et al., 2017). These cows also produce 0.7kg/day more milk than non-shaded cows (Fournel et al., 2017). Furthermore, conception rates in shaded cows were greater (25.3% vs. 44.4%) (Fournel et al., 2017).

Record high temperatures in the United States, especially in southern states, frequently exceed the zone of thermal comfort for dairy cows (St-Pierre et al., 2003). Earlier research predicted the effects of global warming on the dairy industry, emphasizing the critical importance, projected to rise to 1.4 % by 2030, with even higher losses expected in certain states due to geographic factors (Gunn et al., 2019). Abatement strategies have been a topic of discussion for over four decades to address these challenges. However, earlier approaches often overlooked future issues that would arise in the 21st century.

Summary

Climatic changes, including rising temperatures, pose ongoing challenges for dairy producers, leading to significant monetary losses. Heat stress remains a pervasive issue affecting milk yield, reproduction, and metabolic processes. While it presents a growing concern, researchers and producers must continue to explore solutions to mitigate its impact. The following chapter delves into a research experiment and background on why glucose monitoring could serve as practical management tool for early detection of these factors.

Chapter 2: Relationships between circulating glucose concentrations and productivity in heat-stressed dairy cattle

Introduction

Summer heat stress causes numerous harmful effects for dairy cattle. Hormones and metabolites involved in glucose metabolism are one aspect of dairy cow physiology altered by exposure to heat stress. Characterizing the etiology and consequences of these alterations is important for all aspects of dairy cow productivity since nutrition and metabolism affect growth, milk production and reproduction.

While small quantities of glucose are absorbed from the diet, the primary source of glucose in ruminant species is hepatic gluconeogenesis (75-90%) (Abbas et al., 2020). After consuming a meal, the microorganisms of the rumen digest the feed and produce VFA like propionate, butyrate, and acetate (Abbas et al., 2020). VFA are then used for glucose synthesis in the liver. It is important to consider that blood glucose concentrations are affected by both the rate of synthesis and rate of uptake. These processes are affected by the supply of glucogenic precursors, hepatic function, and hormones such as insulin (De Koster & Opsomer, 2013).

Glucose uptake by the tissues is mediated by two different processes: cotransport and facilitated diffusion. Within the lactating dairy cow, the tissues that consume the most glucose are the udder, skeletal muscle, and gravid uterus (when present) (De Koster & Opsomer, 2013). The most common form of glucose uptake is through the process of facilitated diffusion via glucose transporters (GLUT) (De Koster & Opsomer, 2013). While there are several types of GLUT molecules, there is only one that is responsible for insulin-stimulated glucose uptake. This is GLUT 4 which stimulates uptake to the heart, skeletal muscle, and adipose tissue (De Koster & Opsomer, 2013).

Insulin is produced by the beta cells that are found within the islets of Langerhans within the pancreas (De Koster & Opsomer, 2013). The regulation of insulin is complex, but insulin production and secretion increase predominantly in response to elevated glucose concentrations. When dairy cows experience heat stress, circulating insulin concentrations generally increase while glucose concentrations decrease compared to thermoneutral (Wheelock et al., 2010). The directions of these changes in insulin and glucose are unexpected since feed intake declines during heat stress. Lower feed intake results in less glucose availability for stimulating insulin secretion, and since dairy cows don't become insulin resistant during heat stress, there is no obvious cause for elevated blood insulin concentrations.

Full characterization of circulating insulin and glucose concentrations of dairy cows exposed to high ambient temperatures would be an informative approach for evaluating cow response to heat stress. Unfortunately, there is not a cow-side method for measuring blood insulin concentrations. Blood glucose concentrations on the other hand can quickly and easily be measured using hand-held glucometers designed the use of humans, making it a viable option for monitoring individual heat stress responses. This study aims to investigate the use of blood glucose as a monitoring tool to determine the severity at which individual cows are experiencing heat stress. This knowledge would allow producers to manage cows more precisely within their herd, with the ultimate goal being to increase productivity.

Materials and Methods

Barn Environment/Operations

This experiment was conducted at the Dairy Science Complex at Kentland Farms in Blacksburg, Virginia. This project and its procedures were approved by the Virginia Tech Institutional Animal Care and Use Committee (IACUC). Lactating Holstein cows assigned to this project (n=53) were housed in one of four pens in a covered free-stall barn. Cows were milked twice daily and fed according to the farm's standard operating procedures.

Cows having experienced clinically normal periparturient periods were enrolled on a rolling basis beginning at 10 days postpartum. Of the 53 cows assigned to this project, 26 were primiparous and 27 were multiparous.

Temperature Data

For the duration of the experiment, ambient temperature and humidity data was collected with the Lascar EasyLog USB Data Loggers. Four data loggers were placed throughout the barn pens to ensure accurate measurement of temperature and humidity. These temperature collectors were placed above the headlocks (n=2) and free stalls (n=2) where there would be no interference by either cows or humans. The data loggers collected ambient temperature in Celsius (°C), humidity (rh%), and dew point (°C). This information was used to calculate the temperature humidity index (THI) in 15-minute intervals for the entirety of each day of the experiment. The formula used for this experiment is THI= T (dry bulb) + (0.36 * T (dew point)) + 41.2 (Gaughan et al., 2012). The THI information was used to categorize the severity of heat stress each day using the threshold of 68 THI, where THI at or above 68 is considered heat-stress-inducing for lactating dairy cows (Zimbelman et al., 2009). Days with 0 hours above 68 THI were considered non-heat-stress days, days with >0 to 8 hours above 68 THI were moderate-heat-stress days, and days with >8 hours above 68 THI were severe-heat-stress days.

Sampling, Measurements and Record Collections

Sampling, measurements, and record collection started Monday, May 22nd, 2023. Cows were restrained either in the headlocks in the freestall barn, in a palpation rail or in a chute. Blood samples were collected from the coccygeal vein into evacuated herparinzed blood tubes (BD Vacutainer, Becton Dickinson, Franklin Lakes, NJ) twice weekly. Blood glucose concentrations were determined immediately using a hand-held glucometer (Abbott Precision Xtra) that was developed for use by humans to monitor blood glucose levels. Rectal temperatures were taken on the same days that blood samples were collected using the Welch Allyn SureTemp Plus Thermometer.

Glucose was collected during this project to determine if each cow was hypoglycemic during heat stress. Cows were categorized as hypoglycemic if they had sustained reduction of blood glucose on severe-heat stress days. Categorization as hypoglycemic or non-hypoglycemic was limited to a subset of cows with 5 or more blood glucose measurements that spanned different THI categories (n=28 cows).

Milk production was collected using VT PCDART software, in which most cow data (milk production, calving, health conditions), is available for research access. Milk production data was obtained from the software for each sampling day, the day before and the day after sampling.

Statistical Analyses

Data were analyzed using the MIXED and CORR procedures of SAS (SAS Institute, Inc, Cary, NC). Independent variables included the main effects of the severity of heat stress (non-, moderate- or severe-heat stress). Cow served as the experimental unit as heat stress response (blood glucose, rectal temperature, and milk production) was measured on an individual basis. For each analysis, eight covariance structures were tested and the most appropriate structure for each variable was selected based upon Akaike's information criterion, Akaike's information criterion with correction and Bayesian information criterion values. In separate analyses, cows were categorized based upon their glycemic status during HS (either did or did not become hypoglycemic) and changes in blood glucose, rectal temperatures and milk production were analyzed based upon that glycemic status. Results are reported as least squares means ± standard errors of the means. Means were separated using the Tukey procedure of SAS. Statistical significance was declared at $P<0.05$ and a tendency for a difference at $0.05<P<0.15$.

Results and Discussion

In this study, the cows that were sampled were in early lactation and exposure to heat stress was reliant on the weather, and therefore, beyond the control of the researchers. Since sampling was conducted during early summer to early fall, cows were ultimately exposed to a variety of weather patterns. When all days of the experimental period were considered (sampling and non-sampling days), cows in this study overall experienced a daily average THI of 68 or greater for roughly 10 hrs. Across the United States, 14.1% of all annual hours is when dairy cows are exposed to heat stress conditions (Becker et al., 2020). Severe bouts of heat stress are particularly prevalent in southeastern states such as Florida and Georgia, which have a more subtropical climate, which explains the higher hour count compared to the state of Virginia (Becker et al., 2020).

As mentioned, rectal temperatures are a physiological indicator of an animal's ability to maintain homeothermy. Rectal temperatures in other studies have also been associated with respiration rates, and respiration rates are sometimes used in place of rectal temperatures as an indication of individual response to heat stress (since counting respiration rates is a non-invasive technique). The literature mentions that an increase of 1 °C in rectal temperatures (38.5 to 39.5) can be indicative of the animal going from normal conditions to heat stress conditions (Neves et al., 2022).

Temperature humidity index values are generally lower in the early morning (0900 hr) compared to early afternoon and into the evening (1500 hr – 2100 hr) (Vizzotto et al., 2015). These trends held true in the current study as well. Thus, in order to ensure that sample and data collection coincided with the hottest part of the day, rectal temperatures and blood samples were collected in the early afternoon (~1430 hr). Rectal temperatures and THI are often negatively correlated with production-related factors, such as milk yield and conception rate (Yan et al., 2021). As expected, rectal temperatures were greater on severe-heat stress days in the current experiment than on non- and moderate- heat stress days (38.94 ± 0.04 vs. 38.68 ± 0.06 vs. 38.56 ± 0.068 for severe-, non-, and moderate-heat stress categories respectively; $P = 0.01$). Rectal temperatures were also positively correlated with the number of hours of heat stress in a day ($r = 0.46$; $P < 0.01$). (Figure 1) These results demonstrate the direct relationship between the severity of heat stress exposure and the cow response measured as rectal temperature.

Reduced milk yield during summer months is arguably the most problematic outcome of heat stress exposure for dairy cattle. As stated previously, both primiparous and multiparous dairy cows mount physiological response to heat stress, many of which are detrimental to overall productivity. Studies have shown that regardless of parity, milk yield decreases when the THI exceeds 68 (Chen et al., 2022; Zimbelman et al., 2009). In the current study, however, milk production did not differ between severe-, moderate- and non-heat stress days. While this was unexpected, it could be due to variations in heat stress conditions (humidity vs temperature ratios to reach at THI of 68) or individual cow sensitivity to heat stress. For example, work conducted by West and co-workers (2003) found that milk production will start to vary around 39.3 to 39.6 °C with THI ranging from 72.0 to 83.6 (Liu et al., 2019). These values are much higher than the threshold determined by Zimbelman and co-workers (2009) and were seldom reached or exceeded in the current trial.

In this experiment, the relationships between milk production, blood glucose concentrations, and THI were analyzed. When all cows were included in the comparison of hypoglycemic to non-hypoglycemic cows, milk production did not differ on non-heat stress days (45.92 ± 3.77 vs 47.98 ± 3.03 kg/d for non-hypoglycemic and hypoglycemic cows, respectively; $P = 0.67$), moderate-heat stress days (46.06 ± 3.94 vs 48.76 ± 3.17 kg/d for non-hypoglycemic and hypoglycemic cows, respectively; $P = 0.60$) or severe-heat stress days (44.57 ± 3.59 vs 47.91 ± 2.89 kg/d for non-hypoglycemic and hypoglycemic cows, respectively; $P = 0.47$). When separated based on parity, primiparous hypoglycemic cows tended to better sustain milk production than primiparous non-hypoglycemic cows (1.47 ± 0.97 vs –0.85 ± 1.13 kg/d, respectively; $P < 0.15$), while multiparous cows did not differ (-1.95 ± 1.14 vs –1.44 ± 0.85 kg/d for non-hypoglycemic and hypoglycemic cows respectively; $P = 0.11$). (Figure 2) This is similar to the findings of Chen and co-workers (2022), where multiparous cows also did not differ in peak milk production in June with an average THI of 66 (Chen et al., 2022). In the current study, the results suggest that primiparous cows may be more sensitive to the combined effects of glycemic status and heat stress. This makes sense since primiparous cows have not yet reached their mature weight, and therefore had the added burden of growth while lactated during heat stress.

In order to gauge the variability in milk production between severe-, moderate-, and non-heat stress days, standard deviations were calculated. This variability did not differ between hypoglycemic and non-hypoglycemic cows (12.17 ± 1.32 vs 10.21 ± 1.06 for non-hypoglycemic and hypoglycemic cows, respectively; $P = 0.26$). Variability of milk production was greater in multiparous compared to primiparous cows, however (8.80 ± 1.18 vs 13.58 ± 1.21 for primiparous and multiparous cows, respectively; $P < 0.01$). This finding would seemingly contradict our previous conclusion that primiparous cows appear to be more sensitive, but this difference is likely driven by the greater overall milk production of multiparous cows.

As with milk production, blood glucose concentrations did not differ between severe-, moderate- and non-heat stress days. Irrespective of the weather, glucose was negatively correlated with milk production on the day of measurement ($r= -0.25$; $P<0.01$) and the day after measurement ($r=-0.23$; $P<0.01$) (Figure 3). While this could simply reflect the use of glucose by the mammary gland, it does not explain why the magnitude of use by one cow would differ from another. Instead, these findings support previous work suggesting that hypoglycemia during heat stress may be a beneficial adaptation rather than a maladaptation (Stewart et al., 2022).

This study aimed to investigate the relationships between glycemic status and milk yield during heat stress. Taken together, the results of this study and previous work (Stewart et al., 2022) suggest that cows with lower blood glucose concentrations are more capable of sustaining milk yield during heat stress. As mentioned, glucose is vitally important for milk production. It is not surprising then, that blood glucose concentrations are related to milk production even during periods of stress.

Conclusions

Overall, these results are an initial indication that hypoglycemia during heat stress is related to milk production and may be particularly important in primiparous cows. More work is needed to define the nature of these relationships (cause vs effect). Measurement of blood glucose concentrations could eventually prove to be a method for predicting an animal's sensitivity to heat stress. Such information could be used in many ways to improve the profitability of a dairy operation. For example, cows could be grouped differently based upon their predicted sensitivity to heat stress, with the more sensitive cows receiving the most aggressive heat abatement strategies. Breeding schedules could be shifted so that the more sensitive cows do not calve in the late spring or summer when heat stress would coincide with peak milk production. This information could also be used for selection for heat-tolerance. Irrespective of its usefulness for on-farm management decisions, characterization of the relationships between blood glucose and milk production during heat stress adds to our understanding of the physiology of the heat-stressed dairy cow.

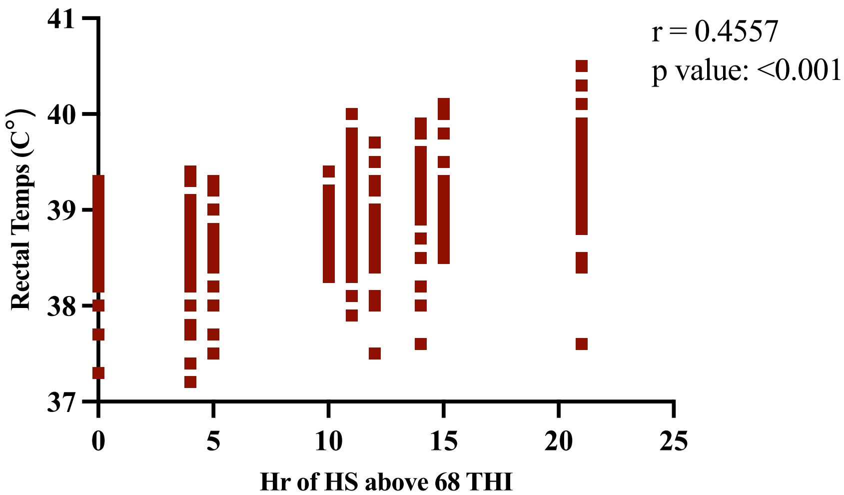

Figure 1. Correlation between hours (Hr) of heat stress (HS) above 68 THI and rectal temperatures.

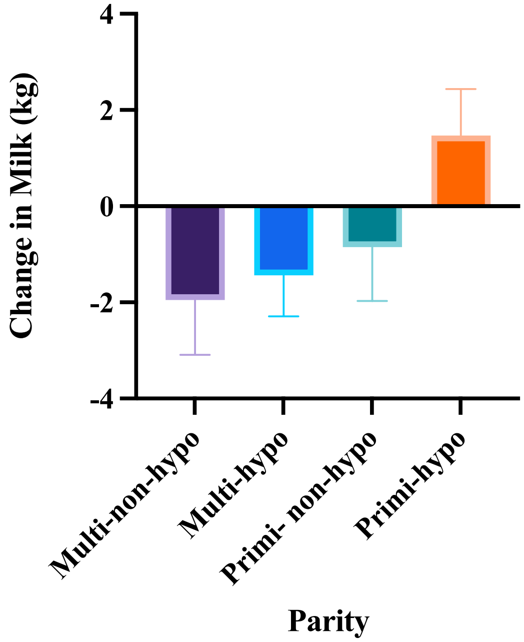

Figure 2. Change in milk (kg) from thermoneutral to heat stress conditions for multiparous cows (Multi) and primiparous cows (Primi) that were categorized as non-hypoglycemic (non-hypo) or hypoglycemic (hypo).

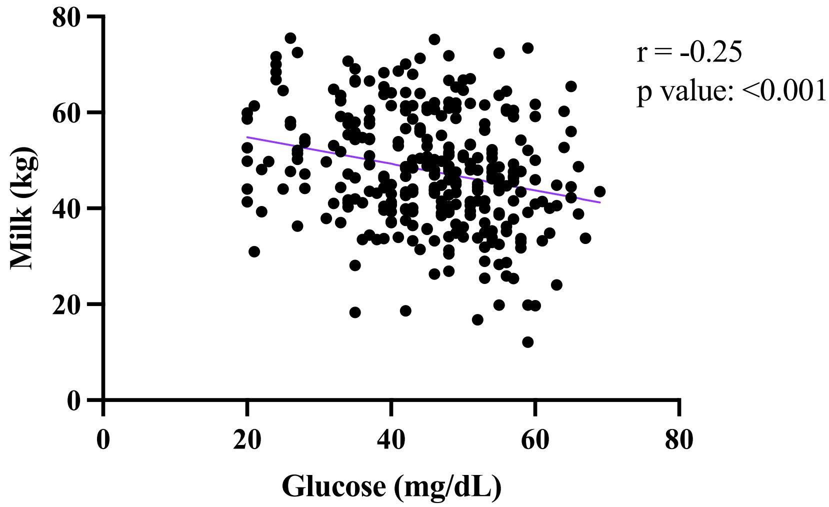

Figure 3. Milk production (kg) compared to blood glucose concentrations (mg/dl).

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