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Heat stress effect on dairy cattle Physiology and selection strategies for thermotolerance

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Abstract

Climate change poses significant environmental challenges that adversely affect the productivity of livestock species. One prominent consequence of climate change is the increased frequency of hot days and heat waves, heightening the risk of heat stress in livestock. Dairy cattle are highly susceptible to heat stress due to their elevated metabolic heat load. Studies have demonstrated that heat stress disrupts numerous physiological processes, leading to substantial economic losses. When faced with heat stress, dairy cattle activate various physiological and cellular mechanisms to dissipate heat and shield cells from damage. These mechanisms demand increased energy allocation towards protection, diverting resources from other biological functions. Consequently, heat stress in dairy cattle results in reduced milk production, impaired reproduction, heightened disease susceptibility, and increased mortality rates. This underscores the importance of selecting thermotolerant dairy cattle. Various strategies have been proposed for conferring thermotolerance, including crossbreeding with thermotolerant breeds, selecting based on physiological traits, and enhancing immune response.

Physiological response of dairy cattle to thermal stress:

Heat stress is characterized as an environmental condition that disrupts the equilibrium between heat accumulation and an animal's capacity to dissipate heat. Increased metabolic heat load generated during milk production makes the dairy cattle highly vulnerable to heat stress. The response to heat stress in cattle manifests through various physiological symptoms as the animal strives to maintain internal homeostasis. When atmospheric temperatures surpass the normal core body temperature, cattle must dissipate heat to uphold normal core temperatures. Consequently, an initial rise in heart rate is observed, facilitating increased blood flow to the body surface. This enables the transfer of heat from the cattle's core to the surrounding environment.

Furthermore, evaporative mechanisms are activated, including increased respiratory rate and sweating. These mechanisms facilitate the removal of heat from the body by augmenting the evaporation of moisture from the skin and respiratory surfaces into the surroundings. Consequently, dairy cattle increase their water intake to counteract water loss through evaporative mechanisms and prevent dehydration. When ambient temperature and relative humidity surpass levels where cattle can effectively dissipate heat, the core temperature of dairy cattle rises above normal, termed hyperthermia. During hyperthermia, cattle reduce their dry matter intake to mitigate internal heat load. However, this reduction in feed intake can lead to deficiencies and complications additionally, alterations in feed composition have been associated with changes in microbial populations.

Heat stress is linked to decreased rumen pH and increased risk of rumen acidosis, attributed to enhanced growth of lactic acid-producing microbes in the rumen during heat stress. Reduced dry matter intake also affects gut villi, causing them to become shorter and broader, compromising barrier function and allowing pathogens to enter the bloodstream through the intestinal lining, triggering an immune response. Consequently, energy is diverted towards sustaining an immune response instead of supporting growth, reproduction, and production. Additionally, various biological processes are shifted to maintain internal homeostasis in dairy cattle. Thyroid hormone levels decrease as a mechanism to reduce metabolic heat load. However, hormones such as prolactin and cortisol increase during acute heat stress, with prolactin potentially influencing sweat gland function and cortisol affecting immune response modulation. During chronic heat stress, cortisol, growth hormone, and thyroxine levels either decrease or remain stable. Elevated levels of progesterone are observed during chronic heat stress, possibly indicating reduced conversion of progesterone to cortisol, resulting in unchanged or decreased cortisol concentrations.

Molecular and cellular processes can be significantly affected by heat stress. Research indicates that heat stress diminishes the gene expression related to transcription, RNA processing, and translation, while augmenting the expression of genes associated with heat shock transcription factor 1 and heat shock proteins (HSP), signalling a shift towards protective mechanisms. Even slight elevations in core temperature can lead to protein misfolding and disrupt the organization of cell organelles, impairing intercellular transport processes. This disruption results in the loss of cellular homeostasis and triggers apoptotic cascades. Consequently, in response to organelle disorganization, HSP expression increases. As internal temperature rises and proteins misfold, bound HSPs are released through the dissociation of heat shock factor 1 monomers from HSP. These monomers then aggregate to form trimers, which are translocated to the nucleus and bind to heat shock elements in the promoter region of heat shock genes. This leads to an increased expression of HSP mRNA, leading

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to the production of inducible HSP. Heat shock proteins serve various protective roles during heat stress; they assist in refolding misfolded or denatured proteins to their native state, prevent aggregation of misfolded proteins, and aid in the degradation of unstable proteins.

Immunological response of dairy cattle to thermal stress:

In addition to impacting productivity, heat stress profoundly affects the health of dairy cattle. Numerous studies have indicated an increased occurrence of diseases in cattle during periods of heat stress. Heat stress can disrupt rumen function and, combined with reduced feed consumption, heightens the risk of metabolic disorders in cattle. Similarly, diseases such as mastitis have been observed to occur more frequently during heat stress, possibly due to enhanced pathogen survival or impaired immune response. Higher mortality rates have also been reported, with mortality rates increasing by 1.27 times during heat waves compared to control periods in dairy cattle. These health-related issues associated with heat stress may stem from various factors, but one of the primary factors could be the impact of heat stress on the immune response.

Both the innate and adaptive immune systems are adversely influenced by heat stress. Heat stress disrupts the adaptive immune response by shifting the balance between T-helper 1 (TH-1) and T-helper 2 (TH-2) responses towards TH-2. Additionally, cortisol, a primary glucocorticoid produced in response to heat stress, inhibits the expression of genes involved in the activation of TH-1 cells and production of pro-inflammatory cytokines by binding to DNA. This leads to impaired cell-mediated immune response, which is primarily responsible for defence against intracellular pathogens. Similarly, heat stress has been shown to down-regulate pro-inflammatory or TH-1 cytokines and up-regulate TH-2 or regulatory cytokines. Collectively, these studies indicate a shift towards a TH-2 response or a suppressive immune response, significantly increasing the risk of diseases caused by intracellular pathogens in dairy cattle.

Heat stress in dairy cattle has been demonstrated to reduce the proliferation of lymphocytes. Lymphocytes, comprising B and T-cells, undergo activation upon pathogen exposure, leading to rapid proliferation to combat infection. Hence, diminished lymphocyte proliferation poses significant challenges for cattle in defending against the pathogens. Similarly, heat stress can impair neutrophil function. This impaired cellular function may result from cell damage or death, as heat stress can increase reactive oxygen species production and reduce antioxidant defences, leading to oxidative stress-induced cell damage. Additionally, heat stress downregulates the expression of L-selectin on neutrophil surfaces, hampering their migration to infection sites.

Furthermore, heat stress impacts the complement system, downregulating genes encoding various complement components, as well as factor B and H. Complement activation enhances

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antibody responses, phagocytic cell function, and pathogen clearance by damaging infected cell membranes. Impaired complement activity hinders cattle's ability to eliminate invading pathogens. Heat stress during late gestation adversely affects the immune system in dairy cattle. Elevated circulating prolactin concentrations, resulting from late gestational heat stress, decrease PRL receptor expression and impair lymphocyte proliferation due to negative feedback mechanisms. Heat-stressed cows during late gestation display reduced oxidative burst and phagocytosis, indicating diminished pathogen uptake and killing, along with reduced specific IgG responses compared to cooled cows. These alterations in immune function expose pregnant dairy cattle to heightened disease risks.

Dairy calves, born with limited immunity, rely on colostrum immunoglobulins (Ig) for protection until their immune system matures. Heat stress during late gestation adversely affects *in utero* calf immune development, leading to increased failure of passive transfer rates and reduced lymphocyte proliferation in calves born to heat-stressed dams. Despite high IgG concentrations in colostrum, *in utero* heat-stressed calves exhibit lower serum IgG levels, suggesting reduced gut absorption capacity. Failure of passive transfer occurs when calves fail to absorb sufficient Ig from colostrum before gut closure, rendering them vulnerable to pathogens.

Selection strategies for physiological traits:

Considering the detrimental effects of heat stress on various physiological and immune functions, it is unsurprising that dairy cattle and calves experience increased disease incidence and mortality during heat stress episodes. Thus, there is a critical need to identify and select animals resilient to environmental stressors amidst current and anticipated challenges posed by climate change. A promising approach to confer thermotolerance in dairy cattle worldwide involves selecting for various physiological traits associated with cooling mechanisms during heat stress. Increased respiration rate and elevated body temperature are among the initial physiological signs of heat stress in dairy cattle. Respiration rate and rectal temperature are considered as potential traits for inclusion in selection programs to enhance thermotolerance. Studies in dairy cattle have reported heritability estimates for rectal temperature ranging from 0.06 to 0.17, indicating the possibility of genetic gains through selection for this trait. However, there are limited studies reporting heritability estimates for respiration rate in dairy cattle, with one study indicating a heritability estimate of 0.04, suggesting slower genetic gains when selecting for this trait. Despite the slow genetic progress in thermotolerance through reduced respiration rate and rectal temperature, the advantage of this approach lies in its minimal impact on milk production.

Furthermore, cellular traits have been associated with thermotolerance, with nitric oxide synthesis being one such trait. Nitric oxide facilitates skin vasodilation during heat stress, aiding in

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heat dissipation. Selecting for enhanced nitric oxide production may improve skin vasodilation during heat stress, enhancing heat dissipation. Another potential avenue for selecting thermotolerance is the investigation of heat shock proteins. These proteins play crucial roles in immune response initiation and cell protection and repair during heat stress. Although heritability estimates for HSP in dairy cattle are lacking, many studies associate HSP with thermotolerance or heat stress resilience. Additionally, increased HSP expression has been linked to reductions in respiration rate and rectal temperature. Therefore, including both physiological and cellular traits in a selection index could enhance overall thermotolerance while minimally affecting milk production and avoiding decreased cold stress resilience.

While selecting for physiological and cellular traits appears promising for enhancing thermotolerance without impacting milk production, the practical implementation faces challenges. These traits are costly and labor-intensive to measure, making it difficult to obtain large datasets. Hence, the scarcity of heritability estimates for physiological traits and the absence of estimates for cellular traits underscore the current infeasibility of including these traits in selection indexes until more cost-effective and efficient methods for data collection are identified.

Selection strategies for high immune response:

The potential trait associated with thermotolerance is immune response. The capability to select dairy cattle with a robust immune response has been developed over several years. The concept originated in the 1970s and 1980s when researchers demonstrated the possibility of identifying mice with enhanced antibody responses to specific antigens. After nine generations of selection, these mice exhibited titers that were 30-fold higher than low responders. High antibody-responding mice also displayed enhanced responses to a wide range of antigens and greater defense against extracellular pathogens. This concept was later extended to poultry species such as chickens, while the simultaneous selection for antibody-mediated immune response (AMIR) and cell-mediated immune response (CMIR) was being evaluated in pigs. It was observed that selecting for both AMIR and CMIR concurrently led to genetic improvements in both traits, resulting in pigs with overall enhanced immune responses. Subsequent studies indicated that pigs bred for high immune responses maintained a balance between type 1 and type 2 responses, while those selectively bred for low immune responses tended to favor a type 1 immune response. These findings suggest that breeding for high immune responses could produce animals capable of effectively combating both extracellular and intracellular pathogens, thereby exhibiting broad-based disease resistance.

The methodology used in pigs to measure immune responses was later adapted for dairy cattle.

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Similar to mice, initial studies demonstrated that dairy cattle could be categorized as high, average, or low antibody responders. Subsequent research incorporated the evaluation of CMIR as well, identifying dairy cattle with high AMIR and CMIR. Numerous heritability estimates have been reported for immune response in dairy cattle over the years, with recent pedigree-based estimates for Holstein cattle reported at 0.45 and 0.18 for AMIR and CMIR, respectively. These estimates indicate moderate to high heritability, suggesting that genes encoding these immune response phenotypes can be passed on to the next generation. Studies have also demonstrated the utility of genomic selection in improving immune response. Significant single-nucleotide polymorphisms associated with both AMIR and CMIR have been identified, with functional properties related to immune response and disease. Combining pedigree-based information on immune response with genomic data enhances selection accuracy and facilitates quicker gains when selecting for high immune response in dairy cattle.

Recent studies indicates that dairy cattle identified as high immune responders may exhibit greater thermotolerance compared to those categorized as average or low responders. Several studies have demonstrated that high immune responding dairy cattle tend to have lower respiration rates at higher Temperature-Humidity Index (THI) values. Additionally, a study evaluating physiological responses to heat stress in a tie-stall facility found that high immune responders generally have lower rectal temperatures during natural heat stress challenges compared to low immune responders. Investigations into the function of blood mononuclear cells during both *in vitro* and *in vivo* heat challenges revealed that high immune responders produce enhanced levels of Heat Shock Protein 70 after multiple heat challenges, compared to average or low responders. Furthermore, high immune responders demonstrated a tendency to produce more nitric oxide over multiple heat challenges in an *in vitro* study. These physiological and cellular traits have been identified as associated with thermotolerance and are potential candidates for selection. Selecting for high immune response in dairy cattle is now relatively simple and cost-effective. With recent evidence suggesting an association between high immune response and thermotolerance, this may prove to be an ideal selection strategy. Thus, offering a solution for dairy farmers who can have thermotolerant animals with uncompromised milk production due to heat stress.

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