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Effect of heat stress on udder health of dairy cows

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Research Reflection

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Abstract

This Research Reflection short review presents an overview of the effects of heat stress on dairy cattle udder health and discusses existing heat stress mitigation strategies for a better understanding and identification of appropriate abatement plans for future stress management. Due to high ambient temperatures with high relative humidity in summer, dairy cows respond by changes of physical, biochemical and biological pathways to neutralize heat stress resulting in decreased production performance and poorer immunity resulting in an increased incidence of intramammary infections (IMI) and a higher somatic cell count (SCC). In vitro studies on bovine polymorphonuclear cells (PMN) suggested that heat stress reduces the phagocytosis capacity and oxidative burst of PMN and alters the expression of apoptotic genes and miRNA which, together with having a negative effect on the immune system, may explain the increased susceptibility to IMI. Although there are limited data regarding the incidence rate of clinical mastitis in many countries or regions, knowledge of SCC at the cow or bulk tank level helps encourage farmers to improve herd health and to develop strategies for infection prevention and cure. Therefore, more research into bulk tank SCC and clinical mastitis rates is needed to explain the effect of heat stress on dairy cow udder health and functions that could be influenced by abatement plans.

Heat stress is one of the most stressful situations for dairy cattle, since the physiological need to balance heat accumulation and dissipation results in a negative effect on overall animal physiology, metabolism and productivity, even leading to death in extreme cases (Mader *et al.*, 2006; Bertipaglia *et al.*, 2007; Vitali *et al.*, 2009; Hansen, 2013; Gonzalez-Rivas *et al.*, 2020). During heat stress, the hypothalamic pituitary adrenal and the sympathetic adrenal medullary axes are recruited to maintain homeostasis (Sejian *et al.*, 2018). Further, cortisol secretion increases linearly with increase of heat stress, leading to immune suppression and increased susceptibility to infections (Ju *et al.*, 2014). The negative effects of heat stress are influenced at cow level by production intensity, genotype, immunity and health status. Accumulation of heat stress is also regulated by key climatic factors including atmospheric temperature, humidity, wind flow and solar radiation (Fig. 1) (Pragna *et al.*, 2017).

The temperature-humidity index (THI) is the most commonly-used heat stress indicator to measure the physical impact of the environment on the dairy cow (Polsky and von Keyserlingk, 2017). Ferreira *et al.* (2016) reported a standard of THI $\geq$ 68 for a heat stress day by calculating National Oceanic and Atmospheric Administration data for each state of the USA. However, due to the distances between the weather stations and farm, significant differences can occur between daily THI recorded in the cow barn and data from the nearest weather station (Shock *et al.*, 2016). Therefore, other physiological factors like rectal or vaginal temperatures, respiration rate, panting, sweating, feed intake, standing time and activity are often used to assess heat stress (West, 2003; Schutz *et al.*, 2008; Hansen, 2013).

Heat stress negatively influences cow health by altering the normal physiological functions of the cow, which result in a higher incidence of udder health problems during summertime (Turk *et al.*, 2015). Moreover, due to high ambient temperatures combined with high relative humidity in summer, the activity of some microorganisms responsible for mammary gland infections is also increased, challenging mammary defence capacity and enhancing bacterial colonization of the gland (Fig. 1) (Barkema *et al.*, 1998; Olde Riekerink *et al.*, 2007, 2008; Fan *et al.*, 2019; Gao *et al.*, 2017). Various time series studies have reported bulk tank SCC above the locally accepted level, e.g. 250 000 cells/ml, in different regions and countries in the world (Lievaart *et al.*, 2007; Kelly *et al.*, 2009; Suriyasathaporn *et al.*, 2012; Hashemzadeh and Khalajzadeh, 2014; DeLong *et al.*, 2017; Aghamohammadi *et al.*, 2018; Macedo *et al.*, 2018). SCC also increases with environmental THI (Nasr and El-Tarabany, 2017), reducing milk value (St-Pierre *et al.*, 2003; Rhoads *et al.*, 2013). Bulk tank SCC is used as a milk payment parameter to improve milk quality in many countries (Barkema *et al.*, 2013; Busanello *et al.*, 2017). In

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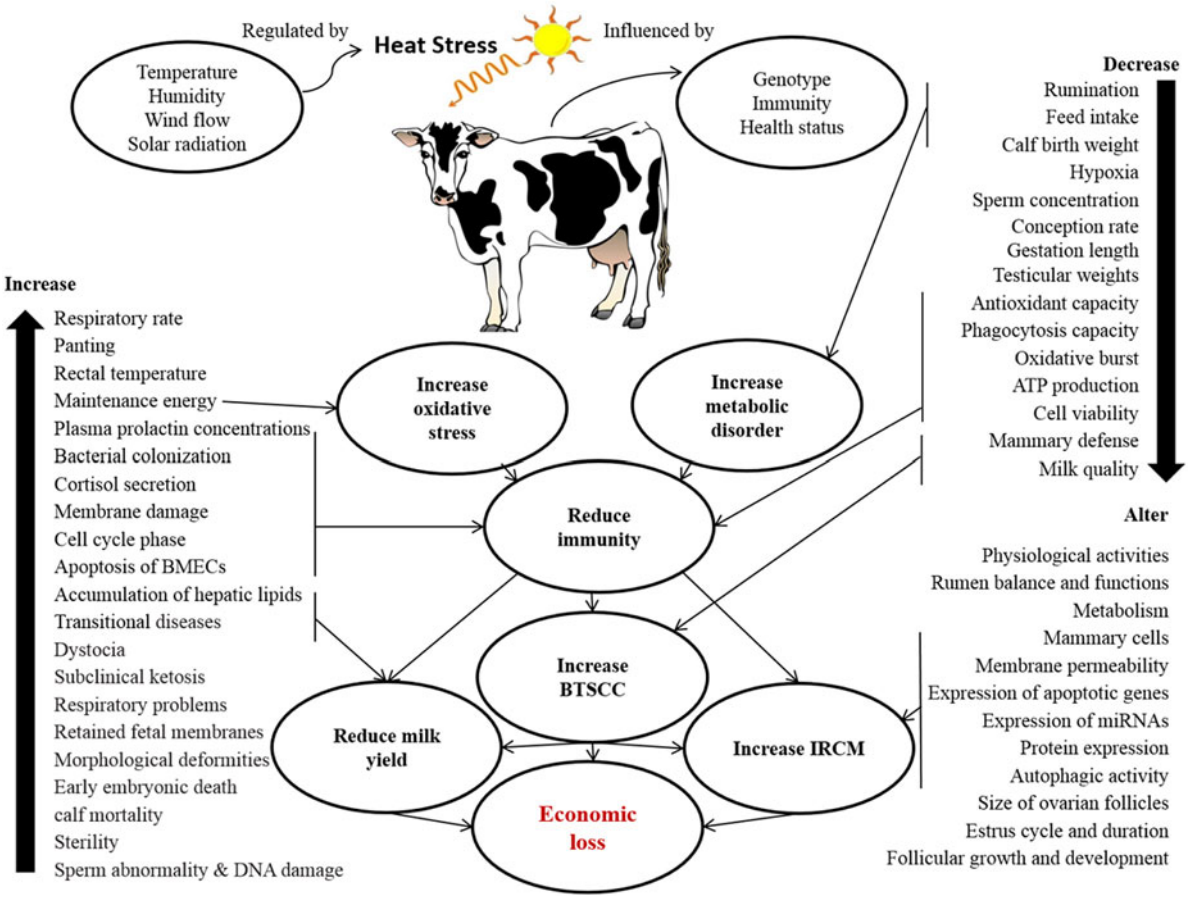

Fig. 1. Various impacts of heat stress on health and productivity of dairy cattle.

Brazil, Lopes and Lima (2018) followed bulk milk samples of 352 dairy farms and reported a negative correlation between SCC and lactose, non-fat dry milk extract, casein and percent of casein in total protein concentrations, conversely, increased SCC is associated with increased contents of total bacterial count, fat and minerals (Lopes and Lima, 2018; Macedo *et al.*, 2018).

The estimated total economic losses due to heat stress in the US dairy industry are more than $800 million annually (St-Pierre *et al.*, 2003; Ferreira *et al.*, 2016) and will probably increase with progressive global climatic change. Therefore, it is essential to better understand the mechanisms through which heat stress adversely affects dairy cows to develop suitable abatement strategies for maintaining optimum mammary gland health during heat stress periods. The purpose of this review is to describe the effect of heat stress on the udder health of dairy cattle, to help develop mitigation plans to maintain udder health.

Heat stress and somatic cell count (SCC)

Bulk tank SCC worldwide

SCC measured at bulk tank level is a widely used indicator when troubleshooting herds with multiple milk quality and allows an estimate of the trends of clinical and subclinical mastitis at the herd and regional level (Olde Riekerink *et al.*, 2007; Rodrigues *et al.*, 2017). Moreover, bulk tank SCC is significantly correlated with polymorphonuclear neutrophilic leukocytes (PMN) content of milk, which is influenced by environmental temperature and act as a vital indicator of dairy herd health (O'Sullivan *et al.*, 1992; Kelly *et al.*, 2000). Although SCC measured at the individual cow level is more suitable for diagnosis and monitoring of mammary gland health, the bulk tank measure is cheap, fast and often preferred by veterinarians and dairy health specialists (Jayarao and Wolfgang, 2003). A bulk tank SCC greater than a target level (e.g. 250 000 cells/ml) often indicates underlying subclinical mastitis (Lievaart *et al.*, 2007; Macedo *et al.*, 2018). Regrettably, and as we have already said, numerous studies have recorded higher levels than this. There are limited time series data regarding bulk tank SCC from Asian and African countries, nevertheless, several studies have found higher values in tropical regions of these continents than in temperate regions of North America and Europe (Kelly *et al.*, 2009; DeLong *et al.*, 2017; Aghamohammadi *et al.*, 2018). Suriyasathaporn *et al.* (2012) in Thailand and Hashemzadeh and Khalajzadeh (2014) in Iran reported bulk tank SCC > 500 000 cells/ml and about 347 000 cells/ml during 2010–12 and 2006–07 respectively. Moreover, Ferreira and De Vries (2015) observed relatively higher bulk tank SCC in milk from hot humid areas of Florida than in lower temperature areas. Whilst not definitive, these various data do suggest possible negative effects of heat stress (Zucali *et al.*, 2011; Smith *et al.*, 2013; Nasr and El-Tarabany, 2017).

Effect of heat stress on bulk tank SCC and incidence of clinical mastitis

The detrimental effects of heat stress on bulk tank SCC and the incidence of clinical mastitis in dairy herds have been reported

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in a number of studies in different regions. Shock *et al.* (2015) assessed the seasonal phenomenon of SCC at the herd level in all dairy farms in Ontario, Canada, over a 24-month period and determined more than 50% of herds experiencing higher bulk tank SCC in the summer months and the next most frequent seasons were fall and spring with approximately 20 and 15% of herds experiencing increased levels in these seasons, respectively. Similar findings were also reported in the USA, Netherlands, Ireland and Thailand (Berry *et al.*, 2006; Olde Riekerink *et al.*, 2007; Rhone *et al.*, 2008; Gillespie *et al.*, 2012) with no significant effect of summer housing on bulk tank SCC (Olde Riekerink *et al.*, 2007). Similarly, it was also reported that SCC increased up to 36% from low to high THI and with a positive linear relationship with the advancement of parities (Nasr and El-Tarabany, 2017).

In addition to the seasonal SCC increase, Ferreira *et al.* (2016) reported lower milk production in the hot and humid summer in Florida with an increase in intramammary infections. They also compared the farm size and observed relatively smaller seasonal effects on bulk tank SCC and milk supply in large farms, which may indicate active commitment to heat abatement. Seasonal variation is also reported in clinical mastitis incidence in several studies with relatively higher incidence in summer months (Salsberg *et al.*, 1984; Green *et al.*, 2006; Olde Riekerink *et al.*, 2007).

Effect of heat stress on mammary pathophysiology

Incidence rate of clinical mastitis in different countries or regions

Clinical mastitis (CM) causes significant economic losses in dairy herds worldwide (Halasa *et al.*, 2007; Nielsen and Emanuelson, 2013) and is regarded as a key indicator of herd health and welfare (Trevisi *et al.*, 2014; Santman-Berends *et al.*, 2015). Among the gram positive bacteria *Staphylococcus aureus*, *Streptococcus uberis* and Coagulase-negative staphylococci (CNS) are the most commonly isolated mastitis-causing pathogens, while *Escherichia coli* and *Klebsiella* spp. are the most often reported gram negative, environmental pathogens (described in online Supplementary Table S1). However, CNS and *S. aureus* were reported more often in North and South American herds than in other continents (Gianneechini *et al.*, 2002; Olde Riekerink *et al.*, 2008; Thompson-Crispi *et al.*, 2013; Levison *et al.*, 2016; Rowbotham and Ruegg, 2016; Tomazi *et al.*, 2018). In China, environmental pathogens like *E. coli* and *Klebsiella* spp. were more commonly associated with CM in summer seasons (Gao *et al.*, 2017). Beyond seasons, environmental pathogens are also linked with housing conditions, hygiene and milking machine management (Bartlett *et al.*, 1992). Contagious pathogens, on the other hand, first attack the udder quarter of the animal, with transmission largely determined by the milking procedure and the milking machine (Barkema *et al.*, 1999).

Beyond these 'mainstream' pathogens, other organisms like *Prototheca* spp., *Arcanobacterium pyogenes*, *Mycoplasma*, *Aerococcus viridans*, *Corynebacterium bovis*, *Trueperella pyogenes*, *Nocardia cyriacigeorgica* and yeast have a considerable impact on clinical mastitis incidence (Liu *et al.*, 2015; Alkasir *et al.*, 2016; Chen *et al.*, 2017; Tomazi *et al.*, 2018; Shahid *et al.*, 2020). In a recent study in southeastern Poland, Jagielski *et al.* (2019) tracked the 4.6% of protothecal mastitis prevalence in individual herds of this region. Shahid *et al.* (2016) and Alkasir *et al.* (2016) identified more *Prototheca zopfii* (13.5%) and *T. pyogenes* (28.6%) isolates (together with other pathogens) from mastitis cases in Chinese Holstein-Friesian dairy herds. Moreover, Verbeke *et al.* (2014) isolated *Prototheca* spp. (1.4%), *C. bovis* (3%) and yeast (2%) from Flemish dairy cattle.

Effect of heat stress on the incidences of clinical mastitis

The seasonal effects on the incidences and pathogen-specific CM in cows were reported in different studies. In Netherlands, clinical mastitis incidence was higher in December to January, whereas incidence of CM caused by *S. uberis* was higher in August, and the incidences of *E. coli* mastitis were higher in summer in confined herds compared with pastured herds (Olde Riekerink *et al.*, 2007). Moreover, in confined herds, environmental pathogens like *E. coli* and *Klebsiella* spp. were more frequently isolated from CM in summer (Makovec and Ruegg, 2003; Gao *et al.*, 2017), suggesting that summer heat and humidity enhance the growth of environmental pathogens. Alhussien *et al.* (2016) also reported higher incidence of mammary infections in indigenous Tharparkar cows in India under heat stress and high humid conditions (>75% humidity). Moreover, in another study in India, Sengar *et al.* (2018) stated significantly higher stress responses in summer seasons for Sahiwal cattle.

Vitali *et al.* (2016) assessed the mastitis prevalence in a large Italian dairy farm over a 24-month period and determined the seasonal factors affecting the occurrence of CM. There was a marked increase during the summer months, with the highest incidence rates at a time when THI exceeded 79, i.e. severe heat stress conditions.

Effect of heat stress on bovine mammary epithelial cells (bMECs)

Under heat stress condition, dairy cows respond with modifications of physical, biochemical and biological pathways to neutralize thermal stress (Baumgard and Rhoads, 2012; Hu *et al.*, 2016). This has a negative effect on antioxidant capacity and immune function, resulting in increased IMI (Almeida *et al.*, 2018; Zou *et al.*, 2019). Heat stress also causes modifications at the molecular level in mammary cells, related to the synthesis of milk components and mammary cell turnover (Salama *et al.*, 2019). Inhibition of cell growth by heat stress results in alterations of structural proteins, membrane permeability and metabolism in bMECs (Salama *et al.*, 2019).

Almeida *et al.* (2018) co-cultured heat stressed bMECs with *S. uberis* and found a dramatical increase in adherence and internalization of this pathogen compared to control (unstressed) cells. This was associated with membrane damage and reduced cell viability. Similar findings were also reported by Kapila *et al.* (2016) in Indian buffalos. Although buffaloes are well adapted to hot and humid climates, they reveal signs of great distress with reducing milk yield and composition, growth rate and fertility when exposed to high ambient temperature. In agreement, Li *et al.* (2015) observed a negative effect of heat stress on cell viability and cell cycle phase by microarray analyses in China. They also provided an overview of gene expression profiles and identified significant alterations of the expression of genes involved in the regulation of cytoskeleton, cellular component morphogenesis, cell cycle, and focal adhesion between heat stressed and normal bMECs. In another study of Li and colleagues, they identified 27 miRNAs which were differentially expressed between the

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mammary tissue of lactating Chinese Holstein cattle in heat stress and regular temperatures. Up- or down-regulation of these differentially expressed miRNAs might be helpful to mitigate the damages due to heat stress (Li *et al*., 2018).

Heat stress also reduces phagocytosis capacity and the oxidative burst of bovine PMN, which together with having the negative effect on the immune system may explain the increased susceptibility to IMI (Lecchi *et al*., 2016). Cai *et al*. (2018) investigated apoptosis of bMECs under heat stress and found a differential expression of apoptotic genes and miRNAs which led to increased apoptosis. However, they also detected an anti-apoptotic effect of miR-216b under heat stress in bMECs by targeting Fas.

Effect of heat stress on dry cows

The transition period from gestation to lactation has significant importance in the production cycle of dairy cows regarding mammary gland development, feed intake and overall production performances of the subsequent lactation (do Amaral *et al*., 2009; Thompson and Dahl, 2012; Fabris *et al*., 2019). However, dry cows often receive lower management intensity since this is seen as a non-productive period (Fabris *et al*., 2019). Dry cows generate less metabolic heat in comparison to lactating cows (West, 2003), which will mitigate the adverse effects of high THI. Nevertheless, heat stress can still have an impact (Tao *et al*., 2011; Ferreira *et al*., 2016; Skibiel *et al*., 2018).

Heat stress significantly affects the dry period of dairy cows by increasing rectal temperature, respiration rate and plasma prolactin concentration as well as decreasing dry matter intake and body weight gain and reducing gestation length and calf birth weight (do Amaral *et al*., 2009, 2011; Tao *et al*., 2011; Wohlgemuth *et al*., 2016; Fabris *et al*., 2017, 2019). In the absence of cooling, heat stress has a negative impact on lymphocyte proliferation in dry cows, and there are indications that the length and intensity of heat stress can compromise immune function (do Amaral *et al*., 2010). Moreover, do Amaral *et al*. (2009) also reported down-regulated hepatic mRNA expression in heat stressed dry cows entering lactation, with greater oxidative burst and phagocytosis in cooled cows (do Amaral *et al*., 2011). They also measured immune status by determination of immunoglobulin G secretion in response to ovalbumin challenge, and suggested that the proper abatement of heat stress during the dry period improved production yield and immunity in the subsequent lactation (do Amaral *et al*., 2011). In a study in Florida, Thompson and Dahl (2012) considered the effects of dry period season on the subsequent lactation regarding production performance, occurrence of health disorders and reproduction performance by including more than 2600 calving records over 3 consecutive years on a commercial dairy farm and reported that cows dried off in hot months had higher occurrence of postpartum disease such as mastitis, respiratory problems, and retained fetal membranes in early lactation compared with cows dried during cool months.

Several recent studies have also examined the effect of heat stress during the dry period on mammary gland development and mastitis control. Heat stress resulted in a decreased rate of mammary cell proliferation, but mammary cell apoptosis was not affected by the prepartum heat stress (Tao *et al*., 2011). Wohlgemuth *et al*. (2016) extended these observations to show perturbations of autophagic activity early in the dry period, and since there are indications that authophagy is required for optimal subsequent proliferation of mammary cells, this may in part explain the negative effects of heat stress.

Skibiel *et al*. (2018) identified differentially expressed proteins by comprehensive bovine liver proteomics analysis of postpartum cows to investigate the protein alterations due to heat stress during the dry period. Hepatic oxidative phosphorylation and mitochondrial dysfunction were the most relevant pathways affected by the dry period heat stress, conversely dry period cooling helped to increase ATP production with reducing oxidative stress, and inhibited the excessive accumulation of hepatic lipids in the liver that might be responsible for fatty liver disease and also provided sufficient metabolic support for higher milk production in the subsequent lactation. Hence, thermal stress management strategies during the dry period may be needed to achieve optimal lactation performance and reduced susceptibility to transitional diseases.

Mitigation of heat stress

Consistent increases of temperature and humidity across the world intensify the dairy industry's need for practical mitigation strategies of heat stress. A number of previous studies have discussed different methods of heat stress mitigation in dairy farms under current or future climatic conditions (St-Pierre *et al*., 2003; Collier *et al*., 2006; Karimi *et al*., 2015; Fournel *et al*., 2017; Bartle *et al*., 2018; Gunn *et al*., 2019). However, successful cooling methods for reducing the effects of heat stress all involve changing the farm environment by maximizing heat exchange through convection, conduction, radiation, and evaporation. Details of abatement strategies vary depending on the farm environment, location, management approaches, stage of production and affordability (Negrón-Pérez *et al*., 2019). Up till now, providing shade to protect against solar radiation is the most effective, simple way of cooling cows (Collier *et al*., 2006; Tucker *et al*., 2008). Veissier *et al*. (2018) evaluated the effect of heat stress on cattle at pasture land in a temperate region of Belgium and reported that providing shade facilities can significantly reduce the negative effect of heat stress on respiratory rates, rectal temperature, milk and fecal cortisol metabolites significantly. Moreover, Collier *et al*. (2006) reviewed the impact of shade and cooling system for heat stress abatement and suggested that the shade area should provide 3.5 to 4.5 m^2 of space for each mature dairy cow and be 4.3 m high to reduce the udder injury and intensity of solar radiation, respectively. Shade orientation should also be taken into consideration during the construction to allow sunlight dispersion beneath the shade.

However, in extreme heat, providing shade alone is insufficient, since it has no impact on environmental temperature or relative humidity. In these circumstances, active cooling is required (St-Pierre *et al*., 2003; Tucker *et al*., 2008; Gunn *et al*., 2019). St-Pierre *et al*. (2003) identified three different models of cooling system using fans and/or forced ventilation, combination of fans and sprinklers, and high-pressure evaporative cooling to abate moderate, high and intense heat respectively in the dairy farm. Fully confined housing offers other opportunities such as earth-air heat exchangers, but these are only really applicable to intensive pig and poultry production (Vitt *et al*., 2017).

Beyond the common management practices for heat stress abatement through active cooling of cows using fans and soaking with water, several studies have shown effects of other factors, like nutritional supplementations, administration of growth hormone and coat color or pigmentation, either alone or in combination

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with active cooling. Growth hormone (recombinant bST) is complicated; it will increase yield even during heat stress but may be associated with reduced fertility, although if management conditions are good then that can be avoided (Jousan *et al.*, 2007). Given that increased yield also means increased metabolic heat, the concept of using rBST to counteract heat stress is questionable, to say the least. Fabris *et al.* (2017) fed a nutritional supplement to achieve immunomodulation in dry cows which, in combination with active cooling, improved milk yield in the subsequent lactation. In Brazil, Lima *et al.* (2019) studied the inclusion of the seaweed *Gracilaria birdiae* in the diet to mitigate dairy goat heat stress and, whilst they found a positive contribution in alleviating stress symptoms, did not see any effect on milk production. Several studies have reported that dietary supplementation with selenium can effectively alleviate heat stress, enhancing immune resistance to oxidative stress and improving feed intake, body weight gain, feed efficiency and productivity in both ruminant and monogastric animals (Calamari *et al.*, 2011; Habibian *et al.*, 2015; Zou *et al.*, 2019).

During summer, dairy farmers also can plan shade management schedule according to the coat color of cattle, because it influences the response of dairy cattle to heat stress as dark coat colored cows can absorb more solar radiation as well as releasing higher rates of heat than light coat colored cattle (Tucker *et al.*, 2008). Moreover, coating the dorsal midline of cattle with a reflective pigment like titanium dioxide has the potential to mitigate the heat stress in feedlot cattle by reflecting solar radiation (Bartle *et al.*, 2018). Despite these positive effects, it is important to remember that exposure of dairy cattle to severe heat stress during summer can have profound negative effects not only on lactation but also on reproductive traits (Jousan *et al.*, 2007; Gernand *et al.*, 2019; Negrón-Pérez *et al.*, 2019).

Conclusions

Heat stress is a distinct physiological condition of dairy cattle, which has a negative impact on many physiological functions including general health, immune function, reproduction and productivity. Although research regarding the direct effect of heat stress on mammary health are limited, the available studies typically describe higher incidences of mastitis and increased SCC during summer seasons compared to winter, and in tropical regions compared to temperate. Heat stress also has negative effects on dry cows, which carry over into the subsequent lactation. Heat stress abatement through environmental management by providing appropriate housing (shade) and cooling (sprinkler systems) can help in recovering optimal cow performance, health and immunity. Complementary nutritional approaches are also available, but it is difficult for a single approach to completely avoid an increased incidence of udder health problems.

Supplementary material. The supplementary material for this article can be found at https://doi.org/10.1017/S0022029920000886

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