

COMBATING HEAT STRESS IN LAYING HENS A REVIEW

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ABSTRACT: Pakistan is located in the subtropical zone of the world where the ambient temperature remains higher most of the year and causes significant economic losses in commercial laying hens. The ambient temperature of some parts in Pakistan reaches up to 52 °C whilst, the ideal temperature for efficient performance for laying hens is 19-22°C. Yet, ambient temperature particularly on the higher side is very disruptive, therefore; heat stress during summer is a major problem of poultry producers in most regions of Pakistan. Several studies have investigated the biohazards of heat stress on production performance, physiology, immunology, digestibility of nutrients, hematology, serology, welfare, behavior and livability of commercial layers. During summer production at farms decreases drastically, thus adversely affecting the economics of layer farms, which might add to a number of culled birds. The key strategies to alleviate heat stress in laying birds are feeding practices and management practices. New innovative approaches such as exploiting and incorporating genes for heat tolerance and genetic marker assisted selection for genotypes with improved heat tolerance should also be explored. Therefore, this review article intends to discuss the effects of heat stress on layer birds' health and production and various management and nutritional approaches to coping with it.

Key words: Heat stress, layers, performance, immunity, digestibility, hematology, serology.

(Received 30.11.2021

Accepted 25.11.2021)

INTRODUCTION

Pakistan is situated in the subtropical zone of the Northern Hemisphere of the world where temperature usually remains well above the higher side of thermo-neutral zone (25-37 °C) for the greater part of the year (Abbas *et al.*, 2021). The ambient temperature of some parts of the region reaches up to 52 °C (Vidal and Walsh, 2010; Abbas *et al.*, 2021; IEE, 2016; Faisal and Riaz, 2019). The optimum temperature for efficient performance is 19-22°C for laying hens, however, ambient temperature especially on the higher side is very disruptive and may reduce survival rate and production (Abbas *et al.*, 2021) performance of layer flock. Therefore, heat stress during summer is a major problem in most parts of Pakistan, and this has pronounced effects on the production performance of commercial laying hens. Egg production declines

drastically, thereby adversely affecting the economics of poultry production, which may lead to an increase in the number of culled birds (Abbas *et al.*, 2021).

Higher temperatures at poultry sheds may cause detrimental effects not only on production performance (egg production, size of egg and egg quality) but also adversely affects the physiology of the birds resulting in high mortality. The birds increase panting up to 10 times if the ambient temperature is higher than thermo-neutral zone. Ambient temperature and circadian rhythm influence liver glycogen and plasma carbohydrate levels with distinct changes in blood glucose (Abbas *et al.*, 2017). As ambient temperature shoots up, respiration of birds increases resulting in greater losses/excretion of CO₂ that cause an increased pH of blood and disturbs the acid-base equilibrium (Abbas *et al.*, 2017) and changes in acid-base equilibrium do cause alkalosis and/or acidosis,

distracting the metabolism used for homeostasis balance rather than used for production. Changes in CO₂ level causes disturbance in pH of blood hence a decline in quality of eggs (Abbas *et al.*, 2017).

Different techniques are being used in poultry production to combat heat stress. These techniques include nutritional amendments such as dietary addition of oils in the diets (Ghazalah *et al.*, 2008), reduction in protein level of feed, supplementation of feed with limiting amino acids and management practices like intermittent feeding, feeding the birds in cool hours of the day, feeding in limited time (Yalcin *et al.*, 2001; Saeed *et al.*, 2019), sprinkling of water, evaporative cooling (Donald, 2000), improved ventilation (Nilipour, 2000) and supplementation of electrolytes (Abbas *et al.*, 2021). These techniques are considered helpful in reducing heat stress. Time limit feeding during cool hours of the day is a general practice for manipulating/alleviating heat stress. It has also been suggested that birds should avoid to feed during hot hour periods (Mahmood *et al.*, 2005) because it only adds to body heat due to heat increment, which the birds has to dissipate. Whereas limited time feeding during the cooler part of the day would increase the feed consumption at a time much suited for its efficient utilization with minimum chances of heat stress. Although, such feed practice is not likely to increase the overall daily feed intake, yet it is expected to improve the feed efficiency and production performance.

The practice of feed withdrawal and intermittent feeding during the hot time of the day is being practiced in many broiler producing areas for the prevention of heat stress and to control mortality. Short term feed withdrawal has been shown to decrease the rectal temperature of birds and increase the capability to stay alive in stressful conditions (Siegel and Jordan, 1997). Practice of supplementation of ascorbic acid in commercial feed has also been considered a useful and effective tool for heat stress amelioration (Khattak *et al.*, 2012). Among the electrolytes, NaHCO₃ may be used to maintain a correct plasma acid-base balance to combat heat stress (Abbas, 2017). Many researchers have reported useful effects of supplementing NaHCO₃ in layers feed during summer (Abbas, 2017; Abbas, 2019; Abbas, 2021). The relationship between production performance and the thermal environment is needed to investigate for more accurate and satisfactory evaluation of quantitative and qualitative performance of laying birds. Scientists tried different techniques to combat heat stress by manipulating the environment and diets of laying birds. The objective of this review is therefore, to investigate and describing the possible devastating effects of acute and chronic heat stress on the production performance of laying hens and different strategies helpful to mitigate the heat stress.

Studies Exploring Adverse Effect of Heat Stress on laying hens

Birds have the ability to maintain the body temperature within a certain narrow range. Higher ambient temperature may cause heat stress which may produce irretrievable thermoregulatory events that are damaging for their life/existence, welfare and performance of birds. This article is an effort to review the research/information concerning the effect of hot weather on laying hens.

What is stress?

Any physical or physiological change in birds due to internal or external deviation in birds caused by an unusual process is called stress (Reddy and Dinesh, 2004). It is an additional burden on the birds, which possibly tends to produce disharmony in various physiological systems. Stress can arise from any of a number of internal or external factors and can cause hormonal changes in the body though pituitary and adrenal glands. General adaptation syndromes described by Selye (1973) has become the basis of studies for many scientists working on the subject of stress in animals. According to him, "stress describes an animal's defense devices, and thus stressor is any condition that provokes protection responses".

The surrounding environment of the bird is amalgamated of interacting stress factors, thus bird's success to compete with it depends on the intensity of stressor and bird's physiological mechanism to react to the stressor (Chrousos and Gold, 1992). Generally the environment of the bird is of two types 1) external *i.e.* temperature, light, etc. and internal *i.e.* disease organisms, parasites etc. Any changes in the environment of birds activate regulatory mechanism in attempt to maintain homeostasis. There are 2 types of regulatory mechanisms: 1) specific, 2) nonspecific (Carrasco and Van de Kar, 2003). Any particular change in the environment of the bird will elude/elicit a specific response. For example, when ambient temperature is increased, it causes: body temperature of bird to rise, vasodilation for rapid/quick heat dissipation, and feathers are rearranged to ease insulation. To regulate adaptation process against stress, endocrine and nervous system work together.

Heat Stress mechanism

During early stages of heat stress sympatho-adrenal medullary (SAM) axis is turned on to regulate the homeostasis. Stimulus of heat stress is received by the central nervous system, which excites hypothalamus to activate pituitary glands (Selye, 1956) and in turn releases Adreno corticotrophic hormone (ACTH). Secretion of this hormone is regulated by the secretion of corticotrophin releasing factor from hypothalamus. Adreno corticotrophic hormone stimulates adrenal gland to produce catecholamine (via adrenal medulla) and corticosteron (via adrenal cortex). Catecholamine (epinephrine and nor-epinephrine) perform non-essential functions *i.e.* increase glucose metabolism (Selye, 1973), blood flow through skeletal muscles (Sahin *et al.*, 2001; 2002), reduce muscle glycogen, oxygen consumption, increase respiration rate,

heart rate, elevates blood pressure and constricts arterioles/venules and increasing neural sensitivity against heat stress. Adrenal cortex hormones (cortisol and aldosterone) perform essential functions *i.e.* promote gluconeogenesis and synthesis of glucose from fat and protein (gluconeogenesis), maintain heat loss mechanism,

blood flow rate and body temperature (Maxwell, 1993; Naga *et al.*, 2018; Abbas *et al.*, 2017). These hormones also maintain osmotic pressure, regulates sodium retention and potassium loss through kidney, water balance and mediate response to stress (Olanrewaju *et al.*, 2006).

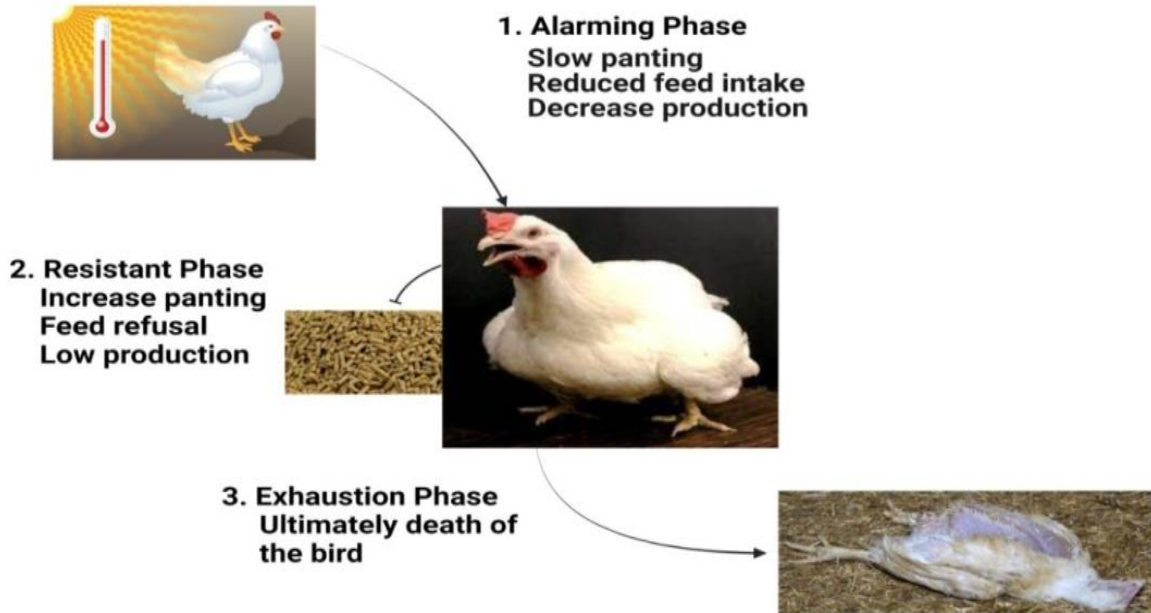


Figure1: Description of three phases of heat stress

Triiodothyronine and thyroxine, also play a pivotal role to maintain metabolism in heat stress birds. Triiodothyronine (T3) concentrations tends to be lowered whilst thyroxine (T4) concentrations was found to be inconsistent in laying hens kept under heat stress conditions (Abbas *et al.*, 2017; Star *et al.*, 2008; Etches *et al.*, 2008). This decrease in T3 concentration might be due to reduction in peripheral deiodination of thyroxine (T4) to Triiodothyronine (T3) during heat stress (Decuypere and Kuhn, 1988). Dwarf gene in *Gallus gallus* found to reduce/prevent the conversion of thyroxine (T4) to

Triiodothyronine (T3) in peripheral tissue (Hoshino *et al.*, 1986). Similarly Triiodothyronine concentration was found to be lowered in Naked neck laying hens than New Hampshire and Lohman white (Melesse *et al.*, 2011). Moreover, plasma concentration of gonadotrophin-releasing hormone level, testosterone, progesterone and estradiol were also found to be impaired/lowered in laying hen exposed to heat-stressed causing reduced production and reproductive performance (Abbas *et al.*, 2017; Nawab *et al.*, 2018; Yoshida *et al.*, 2011; Rozenboim *et al.*, 2007).

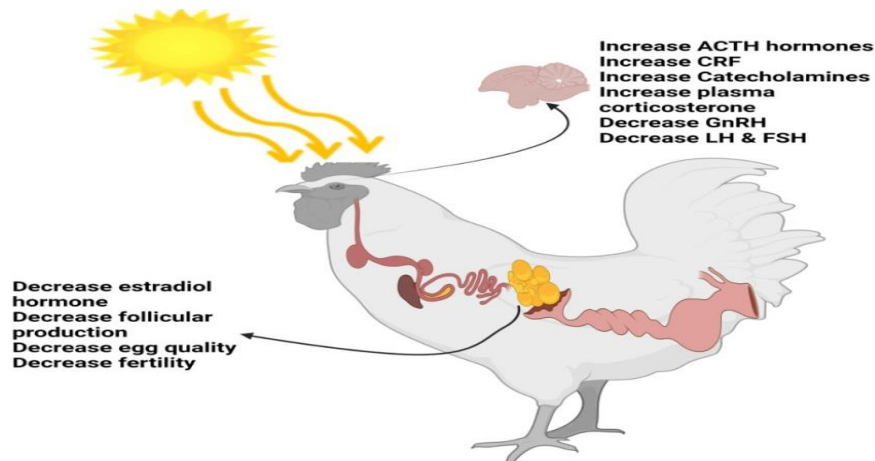


Figure2: Effect of heat stress on Neuroendocrine system of laying hens

Stressors activate a compound array of responses *i.e.* endocrine system, nervous system, immune system etc. and the process is called stress response (Carrasco and Van de Kar, 2003). It causes a number of behavioral and physiological modifications that get better animal existence when there are homeostatic challenges (Abbas *et al.*, 2021). Behavioral adaptations for a stress response may involve increased awareness, euphoria, improved

cognition (Chrousos and Gold, 1992) and enhanced analgesia (Charmandari *et al.*, 2005). Physiological effects due to stress include: increase in cardiac activity, increased respiration, increased intermediate metabolism and restraining of general somatic functions *i.e.* feeding, digestive, growth, reproductive and immune response (Abbas *et al.*, 2019; Abbas *et al.*, 2021).

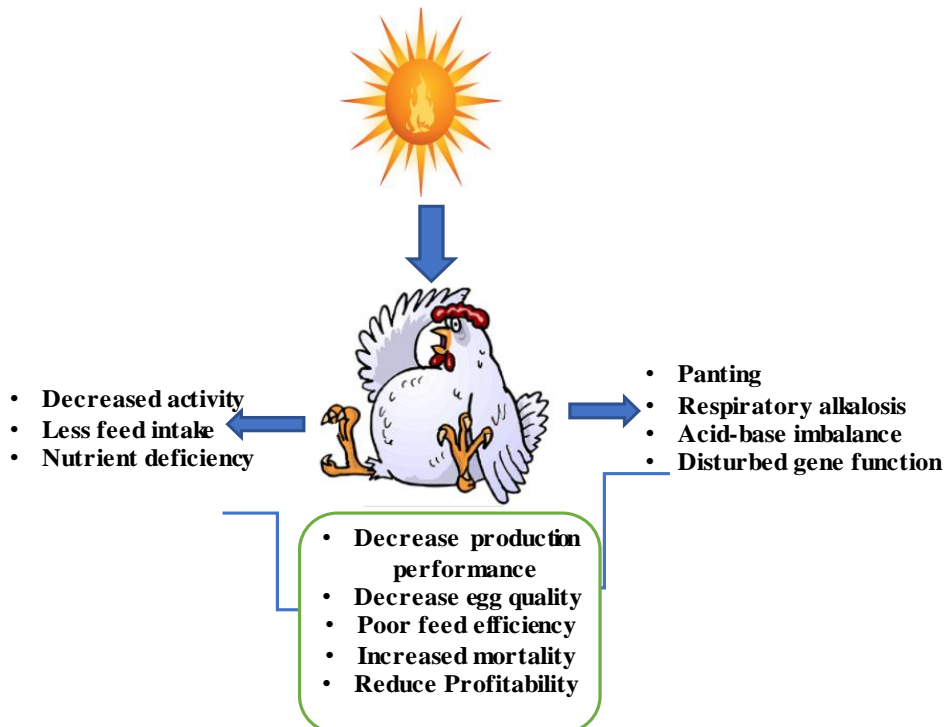


Figure 3: Poultry response to heat stress

Thermoregulation mechanism in birds

Birds are differed in thermoregulation ability immediately after hatching and are categorized as Precocial and/or altricial. Precocial (nidifugous) birds are with down mature and movable from the birth or hatching (Edward and Brinkley, 2000). These are born with open eyes and hatched covered with well-developed downs feather *i.e.* chicken, shorebirds, quails, ducks, geese, rails, coots, waders, tinamous, hoatzin and allies. Precocial birds are capable to immediately respond to temperature changes in the environment thus can withstand fluctuations in their body temperature without damages, however the thermoregulatory ability increases with the age. Precocial birds find their own food, sometimes with help or instruction from the parents. Precocial species are normally nidifugous, meaning that they leave the nest shortly after birth or hatching. The ontogeny of the physiology of control systems is very important for an animal to adapt through life. Generally, thermoregulation process in precocial birds it characterized by three stages.

Thermoregulatory system may work during prenatal phase, however, the efficiency of function is lower and are involved in epigenetic adaptation and get mature in early postnatal stage, the system matures. Full blown homoeothermic stage starts at about the tenth day of age which is described by activation of control elements of thermoregulatory mechanism as well as chemical thermoregulation. Adaptation occurs at genetic, phenotypic and epigenetic levels (Nichelmann, 1992). Genetic adaptation summarizes all morphological, physiological and biochemical peculiarity of the genetically fixed adaptation of an animal to its environment. It is a gross adaptation to the environment. Phenotypic adaptation is innate and caused by changes in gene expression. It occurs during the lifetime and alters the control system and various morphological characteristics of individuals (Tzschentke *et al.*, 2001).

The opposite developmental strategy is called altricial, under which the young are hatched naked and helpless *i.e.* doves, woodpeckers and song birds. Young of

altricial birds are unable to thermo-regulate themselves and heavily depends upon their parents to keep them warm for certain period of time. In most of the vertebrates rapid growth is associated with vulnerable condition during young stage (Case 1978). Avian species whose young develop in exposed nest sites undergo early mobility and early thermoregulatory development (Finch 1984). Altricial avian species particularly are vulnerable during their nestling period and entirely rely on parental for their food, care, protection and to keep them warm. The ability to maintain increased body temperatures is correlated to enabling efficient physiological responses, co-ordinate movements needed for locomotion and feeding. The energy in thermoregulation invested at the cost of tissue development and growth rates (Ricklefs *et al.* 1998; Hohtola and Visser 1998). Environmental pressure i.e. weather, predation, nest sites, food supply, foraging modes etc. determine the growth rates (Ricklefs 1983).

Poultry birds physiologically perform most competently within a narrow comfort zone of 25-37°C (Sturkie, 1976). This range of ambient temperature depends upon body weight of birds, amount of plumage, shape of feathers along with their amount and distribution in body, acclimatization and dehydration status of birds. The higher and lower temperatures are known as upper and lower critical temperatures. A disturbance in ambient temperature disturbs the metabolism and physiological responses including body temperature, diseases, metabolic conditions and production losses (Abbas *et al.*, 2017).

The birds being homeothermic can regulate their deep body temperature within certain narrow limits of ambient temperature i.e. less than 1.8 °F or 1 °C (Abbas *et al.*, 2021). Birds are also endothermic and can rise the rectal temperature by creating significant quantity of heat within the cells and tissue. Birds use plumage, fat insulation and salt glands to regulate their body temperature (Sturkie, 1976). Thermo-neutrality is condition when birds do not use or lose energy to maintain body temperature. When air temperature goes below 16.6 °C heat production is increased in the body. Involuntary muscular contraction and shivering produces heat. However, if temperature drops to a very lower side heat production is not maintained and persistent condition may lead to death of the birds. Birds having more fat deposits can withstand a lower temperature more than birds having less fat accumulation. Various birds have to live merely in certain habitations or migrate among habitats because of physiological limits with respect to environmental temperature. Birds adopt certain behaviors to assist thermoregulation in cold weather i.e. migration to a warm climate, hunch down, sitting (avoid 20-50% of heat loss), huddling together, burrowing, reducing surface area by tucking the head or legs under the wing/feathers to trap heat. Moreover, some birds are able to drop their body temperature (torpid) to endure shortage of food supply in very cold environment.

When air temperature goes yonder thermo-neutral zone (above 27.5 °C for chicken and/or 33-38° C for various other avian species) biochemical reaction speeds up, heat is produced and their body temperature increases (North and Bell, 1990). In order to keep body temperature normal, the birds make effort to dissipate excessive heat via conduction, convection, radiation and evaporation. During humid and hot weather evaporative cooling (panting) is a main source of heat excretion/loss which speeds up water to evaporate through respiratory tract resulting in removal of heat from the body (Angiletta *et al.*, 2010). This mechanism is also affected by ambient relative humidity. Evaporative cooling, fogging and misting of birds are useful even in hot and humid climates. As there are no sweat glands in birds hence they excrete heat through respiration (Abbas *et al.*, 2017). Normally, when atmospheric humidity is increased, the humid environment slows down the vaporization of water molecules from the lungs surface. In this situation birds require more energy to lose heat from body which may cause exhaustion, resulting in heat prostration (Remus, 2001). Birds show behavioral response against higher ambient temperature by avoiding the sun, splashing water over wattle and comb, spreading wings to shade the feet. Signs of heat stress includes, open mouth breathing, wings lifting and gular flutter (account for 35%). Actually mortality is faster and higher due to higher temperature side (heat stress) than cold stress.

Adverse effect of heat stress on production performance of laying birds

1-Growth

Adversative effect of higher house temperature (heat stress) on the production efficiency of laying pullets and broiler is well known (Sohail *et al.*, 2012). There are well documented evidence that growth rate of birds varies with the fluctuations in environmental temperatures. The impact of different environmental temperature on weight gain of birds may also differ dependent on the age and breed of birds (Diarra and Tabuciri, 2014). At high ambient temperature and humidity, the efficiency of chickens to maintain their normal body weight is adversely affected. High ambient temperature ranging 34 to 37.8 °C has shown to reduce body weight gain in chickens (Geraert *et al.*, 1996) and quails (Keskin and Durgun, 1997). Similarly adverse effect of heat stress on body weight gain in birds has also been reported by Abbas *et al.* (2021). Anjum (2000) observed a significant loss in the weight of chicken with gradual increase in ambient temperature. The study of Abbas *et al.* (2021) indicated that layer birds lost significant body weight during higher ambient temperature. They also reported that the seasonal variations affected the rate of daily protein requirements.

2-Feed consumption

Heat stress is known to cause noteworthy

decrease in feed consumption in broilers (Abbas *et al.*, 2021). At higher ambient temperatures body activities of birds are reduced and the bird's needs for energy to keep their body temperature normal are increased which adversely affect the production performance and feed efficiency of the layers (Anjum, 2000). Heat stress is reported to depress the feed intake of chickens (Tadtiyanant *et al.*, 1991). Abbas *et al.*, 2021 studied the significant adverse effects of thermal stress on feed consumption of layer birds. Balnave and Muheereza (1997) observed the effects of high environment temperature (30 °C and 35 °C) on performance of laying birds. They found significantly decreased feed consumption in birds exposed to higher temperature. Mack *et al.* (2013) observed that the birds kept under high environment temperature spent more time in drinking, resting, wings elevating and panting whereas spent less time in feed consuming, moving and walking activities. Feed consumption was reported to be depressed at high ambient temperature (Abbas *et al.*, 2021; Mikec *et al.*, 1992; Muiruri and Harrison, 1991). Similarly, laying hens trained for two meals per day showed decreased feed intake at higher temperature (Li *et al.*, 1992).

3-Feed efficiency

It is observed that heat stress adversely affects feed efficiency of poultry birds in general (Abbas *et al.*, 2021). Poor feed efficiency of the poultry birds may perhaps be ascribed to the reduction in feed consumption of the birds under heat stress. Under these circumstances, a major part of feed consumed is utilized to fulfil the maintenance requirements of the birds. Moreover, a substantial part of the energy is utilized to dissipate heat from their body. Anjum (2000) reported poor feed conversion efficiency in White Leghorn layers exposed to heat stress. Similarly, Li *et al.* (1986) reported poor efficiency of feed utilization in birds exposed to heat stress. However, Muiruri and Harrison (1991) did not observe the effect of environmental temperature on feed utilization efficiency in layers.

4-Egg laying potential

A decline in egg production of layers due to heat stress conditions have been described by various scientists (Muiruri and Harrison, 1991). Anjum (2000) reported that the production performance of White Leghorn birds reduced severely when they were exposed to an abrupt increase in their environmental temperature.

In contrast to the findings discussed above, no effect due to heat stress has been observed on egg production. Roland *et al.* (1996) checked the dietary manipulation of calcium and environment temperature on first cycle egg production of commercial Leghorn layers. Based upon the results of research experiment, warm and cool environment temperatures did not exert any effect on egg laying potential. Heat stress has shown a noteworthy effect on the size of eggs in laying birds and has been

observed to decrease with increase in ambient temperature. Heat stress (Hyperthermia) depressed egg weights in hens (Odom *et al.*, 1985). However, low ambient temperature has been reported to increase egg weight of commercial Leghorn pullets housed at 15.6 °C to 23.3 °C (Ronald *et al.*, 1996).

5-Egg characteristics

It is observed that layer flock kept at higher environmental temperature produced eggs with poor shell thickness (Abbas *et al.*, 2021). Higher environmental temperature has been shown to decrease shell thickness of eggs because of low feed intake (Balnave and Muheereza, 1997), which probably caused reduction in calcium intake, an important element required for shell formation (Abbas *et al.*, 2021). Another probable reason advocated regarding this aspect is decline in calcium level in blood serum due to higher house temperature (Abbas *et al.*, 2017). Anjum (2000) has also reported adverse effect of higher environment temperature on egg shell thickness of the eggs produced by White Leghorn layers and reported that birds exposed to heat stress laid thin shelled eggs.

However, Li *et al.* (1986) have not observed any influence of high environmental temperature on egg shell thickness. Slinger (1985) stated that egg shell thickness decreased when body temperature rose above normal. Deaton *et al.* (1982) determined the effects of high environmental temperature on layer birds acclimatized (adapted) to cyclic thermal environment as opposed to constant ambient temperature. They conducted two trials of eight weeks duration; each on 231 Dekalb layers acclimated (adapted) to twenty four hours linear increase of ambient temperature of either, 15.6 to 35 °C and/or a constant ambient temperature of 39 °C with a relative humidity (RH%) of 26%. They observed insignificant effects on shell thickness of the eggs of layer birds exposed to cyclic vs. (versus) constant temperatures. Akram *et al.* (1999) reported that surface wetting of layers by water spraying can improve thickness of the eggs produced by the birds.

North and Bell (1990) checked the effect of high house temperature on eggs quality characteristics of hen and stated that quality of albumen and yolk deteriorates with the increase in storage time and temperature. A severe problem related to egg production is the appearance of blood and meat spots in the eggs which lowers quality as well as age of the eggs (shelf life). Higher protein percentage in diet is positively correlated with the blood spot occurrence in laying hens. Nair and Elizabeth (1983) reported that the percentage of eggs with blood spots was 4.5-9.5 in the various seasons and that of meat spots was 9.5-15.5; the differences between seasons being significant.

Trail (1963) compared blood and meat spot problem in local chicken breeds of Uganda with five imported ones. The Indigenous breed was found to have

less (0.7%) eggs having blood and meat spots. Whereas, imported breeds were found to have more (2.1% to 9.1%) eggs experiencing blood and meat spots. This difference in the rate of eggs meat and blood spots was attributed to less

stress and good adaptation of hot environment rate experienced by the birds. Akram *et al.* (1999) found less blood spots in restricted feeding of hens under normal laying process.

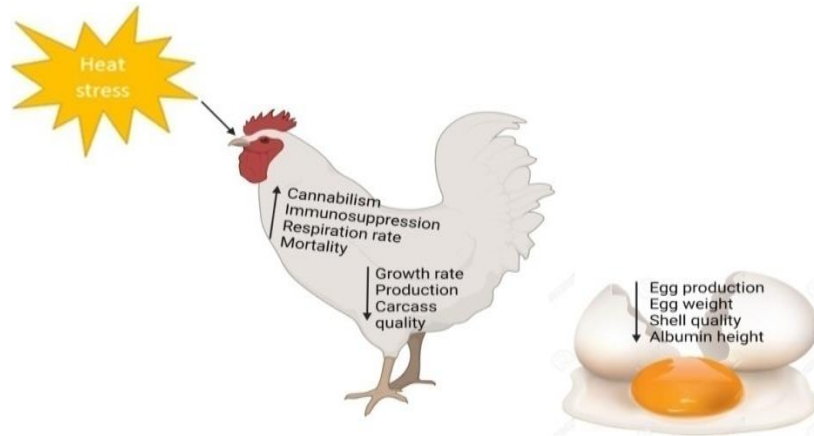


Figure 4: Effect of heat stress on production performance of poultry

Effect of hot weather on Physiological response of layer birds

Among the factors affecting rectal temperature, important ones are humidity, heat loss rate from the body, feed intake, nature of feed, water intake feeding schedules, vitamins, minerals, acids-base balance, electrolytes, breed, strain, housing conditions and cooling methods. Body temperature of adult chickens also differs with change in the environmental temperature (Reddy and Dinesh, 2004). When the ambient temperature is equal to the rectal temperature, heat cannot be lost from the bird by non-evaporative means. However, excess heat get lost through the respiratory lining by evaporation of moisture (Abbas *et al.*, 2017). Birds have air sacs for exchange of heat between the body and the external environment. These air sacs contribute to increase the surface area for exchange of gases and evaporative heat loss (Fedde *et al.*, 1998).

Environmental temperature causes a significant impact on physiological phenomena in birds. Mack *et al.* (2013) reported that birds exposed to higher ambient temperature (heat stress) conditions spent more time for panting. Heat stress disrupts reproductive hormones of layer birds secreted by the hypothalamus and ovary (Elnagar *et al.*, 2010). A decrease in volume of semen fluid, sperm cells concentration and live sperm cells count was observed in males broiler breeders subjected to high temperature environment (McDaniel *et al.*, 2004).

Prieto *et al.* (2010) observed significant less circulating lymphocytes count but an increase in the number of heterophils count of birds reared under heat stress. Moreover, broilers subjected to high ambient temperature showed regressed lymphoid organs weights (Niu *et al.*, 2009), ultimately leading to a poor immune response in laying birds. Birds kept under heat stress showed depressed systemic humoral immune response,

less number of intraepithelial lymphocytes and Immuno globulin A secretion cells in their digestive system (Niu *et al.*, 2009). The reduction in immune response of the birds was attributed to a decreased antibody response and reduced phagocytic capability of macrophages exposed to high ambient temperature. Teeter and Belay (1996) recommended fasting and starvation as a method of lowering the rectal temperature in broilers kept under hot weather conditions.

Effect of hot weather (heat stress) on Hematological profile of layer birds

It is generally agreed that environmental temperature, age, season, feed, diurnal effect, and fasting are related to haematological responses in poultry birds. Thermal environment has shown a significant effect on white blood cells count. Scientists examined the effects of hot weather environment on differential leucocyte (DLC) responses in broilers, turkeys and ducks exposed to various degrees of feed restriction. It was also reported that a mild to moderate heat stress may result in increase in leucocyte count (Maxwell *et al.*, 1992). An increased white blood cells count in laying birds kept at hot climatic conditions is also reported by Anjum (2000).

Lowered house temperature (8 °C) may cause an increase in the hematocrite value, whereas higher house temperature (30 °C) results in a decrease in hematocrit concentration. Higher house temperature exhibits a reduction in packed cell volume in chicken and turkeys (Parker and Boone, 1976). However, supplementation of anti-stress compound like vitamin C and sodium bicarbonate is testified to be effective to improve the PCV (packed cell volume) value in layers exposed to heat stress. Sahota and Gilani (1995) reported the effects of supplementation of ascorbic acid in the diet layers kept

under high ambient temperature and concluded that influence of ascorbic acid on packed cell volume might be due to the rise in erythrocyte number.

Abbas *et al.* (2017) reported an increased red blood cells concentration in birds fed diets containing NaHCO₃. While, Khattak *et al.* (2012) investigated an increased white blood cell concentration in the birds reared under hot climatic conditions than those fed NaHCO₃ containing ration/feed at the same temperature. Oladele *et al.* (2001) reported a note-worthy reduction in PCV in birds kept under higher ambient temperature. This increase was ascribed to high ambient temperature which might impaired the synthesis of blood cells in these birds. Heat stress increased blood glucose and decreased liver glycogen levels in pigeons (Chakraborty and Sadhu, 1983). Abbas *et al.* (2021) concluded that an increase in ambient temperature above the optimum level caused a considerable drop in serum glucose level in birds. Whereas, opposite to it, Yang *et al.* (1992) observed highest blood sugar contents at 23 and 28 °C (223.6 and 221.7mg/100ml) at the exposure of broilers to 12, 18, 23, 28 and 32 °C temperatures.

Al-Hassani *et al.* (2001) have observed an increase in hematocrit values in birds treated with NaHCO₃. They attributed this increase to high house temperature that may hurt the production of red blood cells in these birds. Likewise, results of an experiment executed by Oladele *et al.* (2001) has reported a significant decrease in blood packed cell volume in birds reared under hot weather conditions when fed sodium bicarbonate supplemented diet.

Effect of hot weather (heat stress) on Serum metabolites and serum protein of laying hens

It is reeled that heat stress has been found to affect their serum metabolites and the effect may be ameliorated by fortifying their diets with different levels of sodium bicarbonate. An increase in serum urea concentration in layers kept at higher ambient temperature when compared to those reared under heat combating systems has been reported by Anjum *et al.* (2000). Whereas, Yang *et al.* (1992) has observed significantly higher serum urea concentration in birds kept at low temperature (12 °C) as compared to those kept at relatively higher ambient temperatures (23 and 28 °C). On the other hand, Kurtoglu *et al.* (2007) investigated a non-significant (P=.094) effect due to dietary supplementation of NaHCO₃ on uric acid concentration in Brown-Nick layers.

A marked decrease in plasma protein (albumin and globulin) concentration has been observed in birds kept under higher ambient temperatures (Anjum, 2000) and the decrease was ascribed to higher ambient temperature (heat stress), which hampered the synthesis of plasma proteins in liver. Higher ambient temperature causes a reduction in blood (serum) protein level in birds (Geraert *et al.*, 1996). Yahav *et al.* (1997) revealed a

decrease in plasma protein concentration at higher temperatures.

There is paucity of information about the response of birds with respect to the serum uric acid fractions during hot summer conditions. However, Yang *et al.* (1992) reported higher serum uric acid level in birds kept at 12 °C than those exposed to 23 °C and 28 °C. Scientific information regarding the effect of dietary addition of NaHCO₃ in the diet on serum alkaline phosphatase in the birds exposed to heat stress is scanty. However, findings of Bogin *et al.* (1981) have depicted that broiler subjected to high environment temperature for two hours showed non-significant effect on their blood serum alkaline phosphatase level.

Effect of heat stress on biological antioxidant system of poultry

Free radicals and peroxides are reactive oxygen species (ROS) which produce within the cells during metabolic processes within the cells i.e. ion transportation, cytokine transcription and immune-modulation etc. Excessive production of ROS within cells gets rid of through physiological detoxification mechanisms. Under thermoneutral conditions transcriptional factor activates additional synthesis of antioxidant molecules to deal with ROS species. However, heat stress decreases the efficacy of the antioxidant defense system leave the cells prone to oxidative stress (Betteridge, 2000; Mishra *et al.*, 2019; Surai *et al.*, 2019; Estévez *et al.*, 2015). Excessive production of free radicals during oxidative stress damage cells components i.e. DNA, proteins and lipids; lower growth rates; economic losses; severe health disorders and biological damages even that a severe oxidative stress may to apoptosis and cell death (Lennon *et al.*, 1991; Estévez *et al.*, 2015).

Effect of higher ambient temperature on Plasma electrolyte and mineral profile of laying hens

As ambient temperature exceeds 30°C, it causes an increase in respiration rate of birds, which might go up to about 10 times more than normal rate (Abbas *et al.*, 2017). It has been observed that birds started panting along with increase in blood pH during high ambient temperature. A mild alkalosis (pH 7.55) develops at a temperature of 35 °C, with no raise in body temperature. Further increase in temperature from 35 °C to 38 °C induces moderate alkalosis, whereas, severe alkalosis (blood pH 7.65) occurs at higher ambient temperature i.e. 41 °C (Leeson and Summer, 2001). Findings of Teeter *et al.* (1985) reported that pH values higher than 7.25, lowered the performance of poultry birds. This increase in blood pH can be prevented by reducing panting and rising water intake in birds (Belay and Teeter, 1993). DEB and blood pH are directly related to each other; at 0 DEB blood pH is always acidic, whilst at 350 DEB it shifts to basic. Acid base balance is also considered to be important in controlling blood pH in fowls, which in turns improves the efficiency

of enzymes and ultimately physiological functions (Patience, 1990). An increase in blood pH has been shown to depress feed intake which ultimately reduced the production performance of birds. Production performance of broiler is higher when pH of blood is optimum (7.28), while a reduction in performance is revealed when pH

value is higher than 7.30 or lesser than 7.20. In verity, this normally narrowed range of blood pH establishes the physiology of body enzymes which is required in expressions of good general health and optimum production of animals (Abbas *et al.*, 2017).

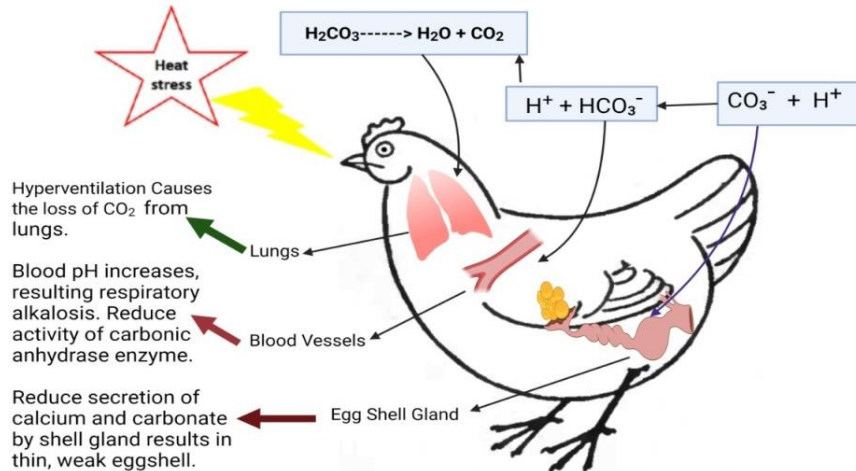


Figure 5: Effect of heat stress on Acid base balance in laying hens

Scientific information about the consequence of dietary addition of $NaHCO_3$ on serum alkaline phosphatase in the birds kept under heat stress is scanty. However, results of some studies designed by Abbas *et al.* (2017) and Bogin *et al.* (1981) have shown that broiler and laying hens subjected to higher ambient temperature showed non-significant effect on their blood serum alkaline phosphatase level. Yang *et al.* (1992) reported higher serum uric acid level in birds reared at 12 °C as than those reared at 23 °C and 28 °C. However, Abbas *et al.* (2017) reported that birds subjected to heat stress shown higher uric acid level in their serum when fed higher dietary level of sodium bicarbonate. Heat stress caused a decrease in the concentration of plasma protein in laying hens with the increase in the environmental temperature (Abbas *et al.*, 2017; Anjum, 2000; Geraert *et al.*, 1996). A reduction in blood Na^+ level in broilers kept under heat stress has been observed by Borges *et al.* (2004). Plasma potassium (K^+) concentration/level was decreased in birds kept in heat stress conditions (Harper *et al.*, 1977) which is reported due to either its increased excretion (Berne and Levy, 1993) or an increase in its uptake by the cells, or both. Increases in excretion of K^+ emerge to preponderate in chronic heat stress, whereas its increase in uptake by the cells is manifested during acute higher ambient temperatures environment. The adverse effect of higher environmental temperature/heat stress on blood K^+ levels is also reported to be alike in various species of birds i.e. layers (Ghorbani and Fayazi, 2009), broilers (Mushtaq *et al.*, 2005), and quail (Keskin and Durgan, 1997).

Effect of heat stress on Serum lipids, hormones and

enzymatic profile of laying hens

Cholesterol is present in the cells of liver and aortic tissues as well as in the fluids of animal body. Important factors which affect cholesterol level in the blood are: sex, age, ration, hyperthermia and starvation. Hevia and Vinsek (1979) reported that fasting can increase the serum cholesterol concentration by metabolizing fats through the gluconeogenesis process hence can finally increase the cholesterol concentration in serum. Higher cholesterol level has also been reported by Sahota *et al.* (1993) and Sahota and Gilani, (1994) in birds kept at high temperatures. Heat stress affects performance as well as various biochemical processes including hormone and enzymatic status of birds (Anjum, 2000). Adrenal gland has a central role in the General Adaption Syndrome (Selye, 1973) and hormones produced by this gland are strongly correlated to the heat stress. Thyroid hormone is vital for the normal growth and development of birds and its secretion rate accurately determines thyroid gland activity (May and Deaton 1986). Thyroid activity was adversely affected by high ambient temperature and it was lowest in chickens reared under heat stress, which might have been due to variations in photoperiod, seasonal reproduction behavior in species and age of the birds (Bowen and Washburn, 1985).

Pituitary and Hypothalamus glands respond to heat stress by decreasing the synthesis of secretions of thyroid gland. Cogburn and Harrison (1980) observed low T_3 values in birds exposed to hot environments. Furthermore, El-Gendy *et al.* (1995) reported a lower plasma T_3 level in heat stressed broilers at 6 weeks of age.

Similar responses on the secretion of T_3 , due to heat stress have also been observed by Brigmon *et al.* (1992) in commercial layers. Moreover, changes in environmental temperature and level of serum T_3 are negatively correlated with each other (Brigmon *et al.*, 1992). Heat stress is recognized to influence reproduction performance in pullets (Abbas *et al.*, 2017; Abbas *et al.*, 2021) and estrogen is a key hormone for efficient reproductive performance in layers. Abbas *et al.* (2017), observed a continuous decrease in plasma levels of estrogen during heat stress. Progesterone is also a vital hormone, which is related to ovulation process of birds. Novero *et al.* (1991) reported that stoppage of progesterone because of malfunction of ovary due to heat stress may cause malfunction of positive feedback mechanism to the hypothalamus resulting in a decrease in the secretion of luteinizing hormone in birds.

Effect of heat stress on immunity level of laying hens

Heat stress is reported to suppress immunity against ND and Gumboroo (IBD) disease in laying hens (Abbas *et al.*, 2019; Mashaly *et al.*, 2004). Environmental stressors affect the immune response and inherent resistance of the host in a direct and/or indirect way (Abbas *et al.*, 2019). Layer birds kept under heat stress experienced a reduction in lymphocytes and a rise in heterophil concentration (Anjum, 2000; Abbas *et al.*, 2017). Thaxton and Siegel (1972) reported that high ambient temperature mediated immune depression. Heat stress caused reduction in total leucocytes and WBCs count and thus affected immune response (Abbas *et al.*, 2017). Environmental factors other than temperature have also shown to influence immunity in birds (Abbas *et al.*, 2017), i.e microbial toxins, hypoxia (Tengerdy, 1970), non-ionic radiation (McRee *et al.*, 1977), social connections (Siegel and Latimer, 1975), heavy metals (Morgan *et al.*, 1975), pesticides (Glick, 1972), nest strain (Thaxton and Briggs, 1972) and amount of nutrients intake (Abbas *et al.*, 2019) etc.

The control of antibody mediated immunity at various environmental temperatures had been studied by many investigators. El-Gendy *et al.* (1995) observed that serum antibodies concentration against Newcastle disease vaccine (NDV) was lesser in heat stressed broilers as compared to those kept under normal temperatures. Birds reared at a temperature of 32.2 °C and higher than 32.2 °C reduced ($P < 0.05$) the agglutinin level in their blood. A petite exposure (2 or 4 times) to cold and subsequent antigen injection increased the agglutinin and hemolytic response in birds. Whereas, 30 minutes cold contact for 2 or 4 times considerably ($P < 0.05$) augmented the IgM antibody concentration and markedly abridged/decreased the IgG (Suba-Rao and Glick, 1977).

Effect of heat stress on nutrient digestibility of laying hens

High ambient temperature may exert a

significant effect on digestibility and absorption of nutrients and their metabolism. Increase in ambient temperature has been shown to reduce feed consumption in birds to prevent thermogenic effect (heat increment) associated with nutrient utilization, absorption and assimilation (Abbas *et al.*, 2019). Higher ambient temperature (above 30 °C) has shown to cause a reduction in blood flow towards gut (Wolfenson, 1986). Resultantly it may reduce hydrolytic activities of the respective enzymes in upper part of digestive tract (Haiet *et al.*, 2000) and hence can cause a decrease in protein digestion. Abbas *et al.* (2027) observed meaningfully lowers protein and carbohydrate digestion in laying hens in which cortisol was dispensed to induce stress. Zuprizal *et al.* (1993) observed a decrease in the digestibility of rapeseed meal and soya bean meal protein, at high ambient temperature. However, Virden *et al.* (2007) investigated that physiological stress had no influence on the digestibility of amino acid.

Factors, which may influence digestibility of nutrients include, ambient temperature (Abbas *et al.*, 2019), level of feed intake and passage rate of digesta (Ravindran *et al.*, 2008), age and physiological status of the bird (Huang *et al.*, 2007), anti-nutritional factors present in feed ingredients (Hughes and Choct, 1999) and nutritional composition of the diet (Leeson and Summer, 2001a). To improve digestibility of feed ingredients different nutritional manipulations have been used such as adding enzymes in feed (Abbas *et al.*, 2021), heat treatment and processing of feed ingredients and addition of electrolytes in feed/water during hot weather (Ravindran *et al.*, 2008).

Attempts to improve feed efficiency of laying hens during hot weather

Heat stress has shown to cause noteworthy ($P < 0.05$) adverse effect on efficiency of feed utilization in birds. Anjum (2000) reported a poor feed conversion ratio in White Leghorn layers exposed to heat stress whereas; Muiruri and Harrison (1991) found that environmental temperature had non-significant effect of on feed efficiency in layers. These types of contradictory findings observed by various scientists are still causing confusion regarding the effect of hot weather on feed efficiency of birds reared under different climatic temperatures, which are direly needed to be addressed.

Several nutritional and managerial manipulations have been used to combat heat stress. These practices include provision of maximum insulation and improving ventilation of the poultry house/shed (Nilipour, 2000), use of evaporative cooling systems (Donald, 2000), thermal conditioning (Yahav, 2000), use of ventilating fans, reducing bird density in the house (Lott, 1991), provision of adequate cool drinking water, feed withdrawal for certain periods or fasting prior to beginning of heat stress, feeding during cool hours of the day and

acclimation (Yamauchi *et al.*, 1995). Al-Zujajy *et al.* (1978) observed that efficiency of feed utilization was significantly better in chicks kept under cool housing conditions.

Use of air cooler improved feed conversion efficiency of the birds by 4.3 to 9.7 percent. Therefore, limited time feeding of birds during cool hours can be a useful practice in poultry birds to combat the heat stress. It has also been suggested that birds should be fed during cool hour periods (Mahmood *et al.*, 2005) because it only adds to body heat due to heat increment, which the birds have to dissipate. Moreover, limited time feeding during the cooler part of the day would increase feed consumption at a time much suited for its efficient utilization with minimum chances of heat prostration. Although, this feed practice is not likely to increase the overall daily feed intake, yet it is expected to improve the feed efficiency and production performance of birds.

Ameliorating/Combating the Heat Stress in laying hens Flocks:

Heat stress severely lessens the profit of poultry farming (Abbas *et al.*, 2017; Abbas *et al.*, 2021). Mitigating heat stress needs multifactorial efforts including genetics (Lin *et al.* 2006), nutrition (Afsharmanesh *et al.*, 2010), thermal conditioning and feeding practices (Abbas, 2017) and housing (Anjum, 2000).

1-Genetic Approach

Recent advances in genetics and biotechnology may lead the way to explore the modifications in genes of chicken to help alleviate heat stress. Developing the

layer lines having incorporated genes that may help to alleviate heat stress in high producers in hot areas is need of time to boost the production for rising human population throughout the globe.

There is a scope of developing naked neck poultry flock in hot areas of the world. Naked neck Gene (single dominant autosomal) helps to lessen feathers in the neck area of chicken, thus facilitate in dissipation of heat through this naked (featherless) area of neck. The gene reduces the feathers covering by 20% in heterozygous and 40% in homozygous (Merat *et al.*, 1986). Naked neck laying hens tend to produce good size and numbers of quality eggs under heat stress as compared to non-naked layers (Fathi *et al.*, 2013).

The frizzle gene create curving outline of the feathers thus helps in reducing weight of feathers and increases dissipation of heat waves (Lin *et al.*, 2006). Homozygous frizzled hens are reported to produce higher numbers of quality eggs as compared to heterozygous frizzled and non-frizzled layers during heat stress. Researches revealed the good effect of frizzled genes on reproductive traits of laying hens but Naked neck genes have more potential to dissipate heat as compared to frizzle genes, however, an additive effect of frizzle gene and Naked neck gene can yield promising results (Zerjal *et al.*, 2013; Yunis and Cahaner, 1999). Dwarf gene is a sex-linked recessive gene responsible for reduction in body weight up to 40% in homozygous males and 30% in homozygous females. However role of dwarf gene against heat stress is need to be explored.

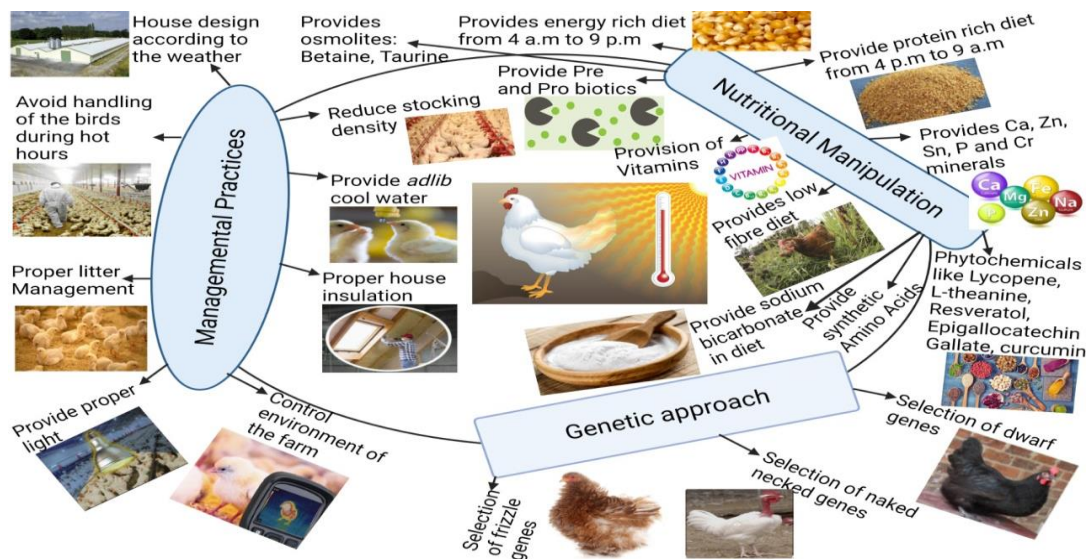


Figure 6: Scientific approaches to combat heat stress in laying hens

2-Nutritional manipulations

A-Lower Protein Diets: Higher protein level in ration increases heat production in the body of birds therefore,

excess intake of protein increases the heat load and disturbs the ionic balance. It is advised that birds should provide a diet of lesser protein concentration which should be supplemented with critical essential amino acids

(Donald and William, 2002) during hot periods (more importantly L-Lysine and DL Methionine).

B-Higher energy rations: Addition of fats/oils in the poultry diets is common practice during heat stress period. Because, fats/oils have lowered heat increment level than proteins and carbohydrates, therefore, some portion of metabolizable energy is obtained by increasing the contents of the diets. Addition of fats help to lessen the specific dynamic effect of rations thus helps to combat the heat stress in poultry birds. Therefore increasing dietary inclusion of fat (up to 5%) poultry rations helps to alleviate heat stress. Higher fat levels of diets also decrease the passage rate of digesta through the gut and increase the nutrient utilization (Attia *et al.*, 2018; Dagher, 2008; Ghazalah *et al.*, 2008). Moreover, Fats/oils have linolenic and linoleic acids (short chain fatty acids), these fatty acids improve weight of the eggs and egg production (Dagher, 2008).

C-Reducing fiber contents of the diet: fibrous materials in moderate levels enhanced performance, nutrient digestibility, immunity affects the gastrointestinal (GIT) development of broiler chicks (Sadeghi *et al.*, 2015; González-Alvarado *et al.*, 2010; Jiménez-Moreno *et al.*, 2009)

D-Adding synthetic amino acids in diet: Improved amino acid balance of the diet can improve the performance of poultry during hot summer (Leeson and Summer, 2001). Formulate to digestible amino acid about 5% to 10 % higher than normal recommendations at the same protein level (Suganya *et al.*, 2015).

E-Use of Betaine in the diet: Betaine has been reported beneficial to ameliorate heat stress in poultry. Betaines act as osmo-protective and maintain cellular water and ion balance (Attia *et al.*, 2016; Leeson and Summer, 2001), works as a methyl donor and can cut the D L methionine and choline requirements of diets involve in fat distribution and enhance immune response. Dietary addition of betaines (0.05–0.20%) has been reported to enabling poultry to resist against heat stress. Effects of Betaine on production performance, physiology, immunity, hormones profile, digestibility and livability of broilers and layers are well documented (Abbas *et al.*, 2019; Attia *et al.* 2005, 2009, 2011, 2016). Taurine (2-aminoethanesulfonic acid) is a most important amino acids present in various tissues of animals. It is important for maintaining calcium homeostasis, bile acid conjugation, membrane stabilization, enhancing thermotolerance and osmoregulation, improving jejunal morphology, and increasing expression of appetite-related genes. Dietary Addition of taurine (0.1% of feed) in laying hen diet may improve oviduct health during heat stress (Dai *et al.*, 2015; Rippes and Shen, 2012).

F-Prebiotic and probiotics: Dietary inclusion of

symbiotic (prebiotics and probiotics) and mannan-oligosaccharides has been reported helpful to mitigate the adverse effect of heat stress in poultry birds (Mohammed *et al.*, 2018; Sohail *et al.*, 2012). Use of prebiotic and the probiotic mixture may be helpful to improve the production performance, immunity and intestinal microarchitecture in birds kept under heat stress conditions (Wong *et al.*, 2006; Silva *et al.*, 2010; Sohail *et al.*, 2011) reported noteworthy improvement in production performance, intestinal health and immunity of birds subjected to heat stress.

G-Supplementation of Vitamins and minerals: Supplementation of vitamins in the feed and/or via drinking water can be helpful to maintain the body temperature of heat stressed birds (Attia *et al.* 2009, 2011, 2016; White head and Keller, 2003; Khattak *et al.*, 2012). Role of ascorbic acid and vitamin E is well documented in this regard. Ascorbic acid (Vitamin C) primarily synthesized by kidneys (Leeson and Summer, 2001) and helps to alleviate oxidative stress in cells and decrease cortisol level and enhances proliferation of B and T cells in birds during heat stress (Attia *et al.*, 2009). Yet, endogenous vitamin C declines during heat stress (Abidin, and Khatoon, 2013). Therefore additional supply of this essential biochemical (250 mg/kg diet) should be checked to improve the performance, immunity, fertility of eggs, hatchability of eggs, reducing oxidative stress and body temperature and improving the carcass grade (Attia *et al.*, 2009, 2011, 2016; Leeson and Summer, 2001; Car *et al.*, 2017). Vitamin E (alpha-tocopherol) works as cell-protective and free radicals scavenger produced in the cells (Attia *et al.*, 2016, 2011, 2009; Khan *et al.*, 2011). Dietary inclusion of vitamin E helps to protect macrophages, lymphocytes and plasma cells against oxidation stress and hence may help to improve performance, immunity, nutrients digestibility, egg production egg quality characteristics and physiology in poultry birds (Abbas *et al.*, 2017, 2019, 2021; Attia *et al.*, 2016). A plentiful intake of alpha-tocopherol (250 mg/kg feed) improves the prostaglandins, leukotrienes and cytokines production. Moreover, combination of alpha-tocopherol (100 mg/kg diet), ascorbic acid (200 mg/kg diet) and probiotics (2 g/kg feed) proved effective to combat heat stress in broilers kept at higher ambient temperature. Vitamin A (Retinol) is an antioxidant of choice at low oxygen tensions which enhances T cell proliferation and antibody production and neutralize thiyl radicals. Dietary inclusion of Retinol (6000-9000 IU/kg of laying hens diet) found to improve the egg weight during heat-stress and to achieve good of antibody titer against ND post vaccination (Palace *et al.*, 1999; Lin *et al.*, 2002). Zinc is coenzyme of more than 300 biochemical reactions and is also part of carbonic anhydrase system which is essential for eggshell formation (Balnave and Muheereza, 1997), hence dietary inclusion of zinc (80–100 mg/kg of layer diet) is helpful to improve

the egg quality of laying hens during heat stress conditions (Balnave *et al.*, 1993). Chromium is an integral part of chromodulin and is needed for the proper function of insulin. Moreover, it is involved in metabolism of proteins, carbohydrates, lipids, and nucleic acids (Vincent, 2000) therefore, dietary inclusion of chromium (0.4–2 mg chromium/kg of die) may results in improved production performance, egg quality immunity and blood profile of laying hens (Torki *et al.*, 2014; Sahin *et al.*, 2002; Li *et al.*, 2001). Selenium is an important component of many seleno-proteins as a par various enzymatic biochemical reactions i.e. thioredoxin reductases and glutathione peroxidase (Nazıroğlu *et al.*, 2012). For example it is essential component of Type I deiodinase enzyme which convert T₄ into active T₃ (Arthur *et al.*, 1992)]. Selenium is reported to enhance the production and reproduction performance of laying hens under heat stress conditions (Attia *et al.*, 2010). Dietary minerals requirement especially calcium and phosphorus increase 4% to 5% however; excesses or deficiencies should be checked at the same time (Leeson and Summer, 2001).

H-Use of phytochemicals/herbal compounds:

Phytochemicals such as Anthocyanins, L-theanine, Lycopene has been tested to relieve heat stress in poultry which has positive effect on health of poultry. These Phytopigments works as antioxidant, immunomodulatory, neuroprotective, anti-stressor, antidiabetic, anti-obesity, anti-inflammatory, and anticancer in nature and have several health benefits like improving performance, egg and meat quality, immunity etc. in birds reared during hot summer (Arain *et al.*, 2018; Sahin *et al.*, 2011; 2013; 2016; Sun *et al.*, 2015; Zhang *et al.*, 2014; Srivastava and Srivastava, 2015; Palozza *et al.*, 2012; Sahin *et al.*, 2011; 2016;). Phytochemicals such as lycopene found in vegetables and fruits like pink grapefruit, papaya, apricots, guava, watermelon and tomatoes (Tanaka *et al.*, 2012) may improve cell growth and modulates phase I and II detoxifying enzymes, activates host antioxidant enzymes i.e. glutathione peroxidase, catalase, superoxide dismutase and others (Martinez *et al.*, 2008), also mediates gene transcription (Palozza *et al.*, 2012). L-theanine (Gamma-Glutamylethylamide), an amino acid found in green tea leaves potent unique health advantages like antioxidant, antidepressant, immune booster (Saeed *et al.*, 2018; Li *et al.*, 2016).

Resveratrol are group of polyphenols reported to enhance antioxidant production and mostly found in peanuts, turmeric, grapes and berries. Dietary supplementation of Resveratrol is helpful to decrease the body temperature, serum cholesterol, corticosterone and adrenocorticotropin levels. Intake of these natural bioactive compounds may cause an increased level of T₃, catalases, glutathione, glutathione peroxidase and superoxide dismutase, in poultry subjected to heat stress

(He *et al.*, 2019; . In laying hens, supplementation of (200 mg resveratrol/kg diet) improve production performance, whilst an inclusion of 400 mg/kg diet can result in reduction the serum/ egg cholesterol and triglycerides (Zhang *et al.*, 2019). Epigallocatechin gallate (EGCG) present in green tea extract are polyphenols having good anti-inflammatory and antioxidant properties reported to improve feed consumption, production performance in laying quails during heat stress (Sahin *et al.*, 2010). Curcumin are animals readily absorbable polyphenols of turmeric having antioxidant/anti-inflammatory properties (Attia *et al.*, 2017) Dietary inclusion of curcumin (150 mg/kg diet) cause an increase in production performance, enhances activity antioxidant enzymes, and improve immunity in laying hens kept under heat stress conditions (Liu *et al.*, 2020)

I-Supplementation of electrolytes: Several methods have been proposed to ameliorate high ambient temperature in the poultry house and to decrease body temperature of birds for successful poultry production. Electrolyte supplementation has been promising to achieve acid-base balance in birds reared in hot summer (Abbas *et al.*, 2021; Leeson and Summer, 2001). Therefore, dietary inclusion and/or water supplementation of electrolytes is a common practice in the field during summer (Abbas, 2017; Akhavan-salamat and Ghasemi, 2016). Dietary electrolyte balance (DEB) in poultry birds plays a significant role for the better performance. An optimum dietary electrolyte balance is thus required for efficient performance, good bone growth and maintaining bedding condition (Oliveira *et al.*, 2010). However, if DEB is not maintained in normal limits, the performance of the birds is adversely affected. Maiorka *et al.* (2004) recommended a dietary DEB of 174mEq/kg for better feed consumption and 163mEq/kg for the best weight gain as compared to 250mEq/kg of DEB. It has also been observed that a DEB of 175mEq/kg may improve performance in broilers until 21 days of age (Szabó *et al.*, 2011), but DEB should be 250mEq/kg during the growing and the finisher stages in broilers. However, role of DEB on the performance of layer birds has not been much studied. Akhavan-salamat and Ghasemi (2016) have reported that, under tropical conditions, using a DEB of 250mEq/Kg achieved a correction of the lay-induced metabolic acidosis and results in a positive effect on performance of layers.

Electrolytes maintain ionic and water balance in living systems. It is important to note that requirements of electrolytes cannot be considered individually because there must be an overall balance among these to achieve homeostasis. Maintaining acid-base balance is a key strategy to avoid harmful effects of heat stress. Acid base balance is mainly affected by environmental and nutritional status of the birds. High anions (negative charged ions *i.e.* Cl⁻) may cause acidemia in chickens, whilst high cation contents (positive charged ions *i.e.* Na⁺,

K⁺) in diet cause alkalemia. Both these adverse situations, therefore, may affect performance of fowls. Dietary electrolyte balance may be calculated using equations developed by various scientists. However, for the calculation of DEB, it be concerned that concentration of sodium (Na⁺), K⁺ and chloride (Cl⁻) should be within adequate range (Abbas *et al.*, 2017). Physiological stress, however, tends to cause deviation in electrolyte balance of poultry birds (Yalcin *et al.*, 2004).

In young birds, Cl⁻ at high levels *i.e.* 160-240mEq/kg, significantly decreased blood H⁺ concentration (Ruiz-Lopez and Austic, 1993). Sodium sulphate has been found to be relatively more acidic as compared to calcium sulphate and potassium sulphate. Gorman and Balnave (1994) investigated that heat stress can cause an increased metabolic need for HCO₃⁻ ions. Patience (1990) observed that acid base and electrolytes balance effect the growth, appetite, thermal stress response and the metabolism of different nutrients in birds. Borges (2001) viewed that a complete electrolyte equation would be expressed as, (Na⁺+K⁺+Ca⁺²+ Mg⁺²) - (Cl⁻ + SO₄⁻² + 2PO₄⁻²) and also reported that maximum feed intake was noted at DEB 264mEq/kg. Rondon *et al.* (2000) reported 250mEq/kg DEB when Na⁺ level were different and 319mEq/kg when K⁺ level manipulate.

Murakami *et al.* (2001) recommended optimal DEB between 246 and 315mEq/kg for broilers during starter phase and for the growers between 249 and 257mEq/kg to achieve efficient performance. Borges *et al.* (2002) investigated that ideal DEB was found to be between 246 and 277mEq/kg. Borges *et al.* (2003) reported that DEB (dietary electrolyte balance) of 240mEq/ kg influenced positive effects on body weight and feed efficiency versus dietary electrolyte balance of 0, 120, and 360mEq/kg, in chicken reared under heat stress. They concluded that an optimum DEB range of 220 mEq/kg to 240mEq/kg be maintained for adequate performance. Barbosa *et al.* (2014) revealed that electrolyte balance may affect intestinal length, water intake and heart and liver relative weights. They concluded that electrolyte balance of 120mEq/kg in feed and 30mEq/L in drinking water may cause an increase in water intake of European quails reared under hot temperature.

Johnson and Karunajeewa (1985) investigated the dietary effect of mineral inclusion *i.e.* calcium and available phosphorus and electrolytes *i.e.* sodium, potassium and chloride on physiological response of chickens. They did not observe any change in plasma ions concentration (Ca, inorganic P, Mg, Na, K and Cl) of birds due to treatments.

J-Dietary inclusion of NaHCO₃: Supplementation of diet and/or water with salts such as NaHCO₃, NaCl, potassium bicarbonate, KCl, and ammonium chloride may increase water intake of birds reared in heat stressed conditions so helpful to maintain electrolyte balance

(Abbas *et al.*, 2021; Abbas *et al.*, 2019; Abbas, 2017; Leeson and Summer, 2001). NaHCO₃ (sodium bicarbonate) is reported to be widely used in poultry feed during summer (Teeter *et al.*, 1985; Abbas *et al.*, 2017, 2019; 2021; Hassan *et al.*, 2009). For normal metabolic events such as maintaining the normal structure and functions of proteins etc., blood pH of poultry birds must be very near to narrow physiological range of 7.35 to 7.45 (Abbas *et al.*, 2017). Moreover, blood pH is closely related to HCO₃⁻ buffering system, which is the major buffering system for maintaining blood pH and can be defined with the following equation.



Blood bicarbonate concentration is primarily under the control of kidneys and to a less extent, the lungs. Kidneys organize the concentration/level of HCO₃⁻ by adjusting its re-absorption from the renal tubules. Increased breathing under heat stress decreases pCO₂ which in turn causes an increase in pH that induces respiratory alkalosis (Belay and Teeter, 1993). The bicarbonate buffer system functions works with double regulatory control of the lungs and kidneys. In HCO₃⁻ buffering system blood pH is represented by the expression (Berney and Levy, 1993) called Henderson Hasselbalch equation as follows.

$$\text{pH} = 6.1 + \log [\text{HCO}_3^-] / 0.03 \text{ pCO}_2$$

Where;

$$\text{pCO}_2 = \text{partial pressure of CO}_2$$

In normal physiological phenomenon the ratio of HCO₃⁻ to pCO₂ is 20:1. In an attempt to keep the body temperature normal, the respiration rate of birds increases which lowers the pCO₂, hence increasing the Log term in the Henderson Hasselbalch equation. This causes an increase in the pH (respiratory alkalosis). In such conditions, NaHCO₃ might be used as a buffering agent to nullify the problem. NaHCO₃ is buffering agent which is helpful to regulate the electrolyte balance and biological pH of the body. Moreover, it is source of CO₂ and can replace the loss of C O₂ occurs due to panting in heat-stressed birds. Layer birds fed diets containing varying levels of NaHCO₃ during summer showed improved performance, digestion of nutrients and immunity against Newcastle disease (Abbas *et al.*, 2017). The use of sodium zeolite and aspirin are also proven to alleviate heat stress.

3-Management Practices

A-House design and location

Location and site of the building, its orientation, plantation and shade, roof color, reflectivity, pitch, R values of construction materials, ventilation system, use of sprinklers and foggers and evaporative cooling system may have a significant effect on heat loss via building. (Moudgal *et al.*,1992; Donald and William, 2002). Designing and orientation of poultry house are important parameters to evade the perception of heat waves inside

the house (Donald and William, 2002). Design of house should be made in such a way that it will achieve maximum insulation so that inside temperature of the house be maintained in thermo neutral zone (Scanes, 2015). Hose must be oriented from East to West in length. The roof of the poultry farm should be insulated, steeper and higher (Donald and William, 2002). Use of sprinklers during heat stress and paint the roof surface with metallic zinc paint and/or installing an aluminum can helpful to cool the building (Donald and William, 2002). During heat stress, evaporative cooling system (through cooling pads, foggers and/or sprinklers) in houses can successfully mitigate devastating effect of heat stress (Attia *et al.* 2006).

B-Light regime: Lighting schedule should be adjusted to offer more light hours during cool times of the day to encourage feed consumption during the cooler hours. A good working stand-by generator must be ready in case of electric short fall during summer.

C-Stocking density: It is advised to reduce stocking density during hot weather. High stocking rate during hot season can result in improper ventilation. Metabolic heat produced by birds and decreased loss of heat due to improper ventilation during heat stress may rise the temperature of the poultry house. Therefore floor space should be adjusted keeping in view intensity of heat stress conditions (Donald and William, 2002). Avoid to use anticoccidial drugs during heat stress.

D-Litter management: Litter caught higher temperature during hot weather (Abreu & Abreu, 2004). Hot and wet weather cause the litter wet. Wet litter increase the growth of pathogens, attracts flies, produces ammonia in house and form cakes resultantly increase the stress upon birds, therefore litter should be maintained well dried. (Donald and William, 2002).

E-Early heat conditioning: EHC (early heat conditioning) also seemed to be one of the favorable method to induce heat adaptation/resistance of poultry breeds to hot weather (Ayo *et al.*, 2010).

F-Feeding practices: Fasting hours lessen the metabolic heat output from digestibility of nutrients, their absorption and metabolism (Richards and Proszkowiec-Weglarz, 2000). Starving/fasting during hot hours also helps to keep calm the birds (Saeed *et al.*, 2019). Feed with drawl two hours before hot hours can increase resistance against heat stress, enhances survival, and reduce heat production and mortality in birds (Yalcin *et al.*, 2003). A controlled feeding schedule can minimize the detrimental effect of hot weather on performance of layer birds. Afternoon or near sunset feeding is also favorable for efficient production during hot weather (Anjum, 2000). Replacing pellets with mash may improve the production performance and water intake in laying birds. Whole grain or coarse mash feeding may increase digestibility during

heat stress. The practice of off feed during hot hours may decrease the heat load and mortality (Yalcin *et al.* 2001). However, physiological adaptation of reduction in feed intake (Abbas *et al.*, 2017) of heat stressed birds causes a reduction in essential nutrient intake (Mahmood *et al.*, 2005; Suganya *et al.*, 2015). Therefore, efforts should be done to stimulate feed intake during hot weather. Another approach is adopting dual feeding regime i.e. provision of high energy diet from 9 a.m. to 4 p.m. and provision of a diet rich in protein from 4 pm. to 9 am may be helpful to maintain the body temperature of heat stressed poultry birds (Lozano *et al.*, 2006). Keeping the feed wet has been reported to be useful reduce viscosity in the gut and increasing water intake during hot summer (Lin *et al.*, 2006). Thus can cause pre-digestion and improve feed intake, dry matter intake, egg production and egg weight (Lin *et al.*, 2006). Cooling effects of fresh water associated with a high intake and extra water intake which improves the performance of layers and broilers (Lin *et al.* 2006; Afsharmanesh *et al.*, 2010.)

G-Ventilation: High air speed is necessary to relieve the birds during heat stress (Nilipour, 2000). In addition, heat loss by convection plus radiation can rise significantly with an increase in air flow (Hilman *et al.*, 1985). In extremes hot and humid weather, evaporative cooling might be ineffective (Attia *et al.* 2006). However, increased ventilation may be helpful to expel hot air from (Dozier *et al.*, 2005). Internal re-circulating fans and exhaust fans can be helpful for uniform circulation of air in the poultry houses aiding convectional heat loss (May *et al.*, 2000). A water level falling through the cooling pads that decreases temperature by 6 °C must increase % RH by about 27%. Therefore, adjusting fan thermostats to run keep house temperature humidity at normal limit (Attia *et al.* 2006).

H-Water: Birds tend to eat less and drink more during hot weather to reduce their body temperature (Abbas, 2017). Normally birds drink twice than what they eat at 15°C. However, during heat stress (30°C - 35°C) the proportion increases i.e. 4.9:1 (Holik, 2010). Provide *adlib* clean and cool water adding ice to water. Increase drinker space and number of drinkers to ensure enough supply of water to cool down the body temperature of birds (Abbas, 2017). To increase water intake during summer use water troughs having sufficient water flow instead of nipple or bell-type drinkers. Place water tank in shade.

I-Health status of birds: Healthy birds can resist the effect of heat stress therefore birds should provide sterile environment during summer to maintain proper health status. For this supplementation of multi-strain probiotics can be used to restore microbial balance and their natural existence. Routinely screening, diagnosis and quick cure of diseased/stressed birds can be helpful to escape the devastating effect of hot weather (Donald and William,

2002).

J-Farm Activities: Loading and transportation of birds is unavoidable practice at poultry farms. However birds should be shifted and transported in special care protocol during transportation and loading in hot summer. Loading, transportation, beak trimming, transfer and vaccinations of birds should be done in cool parts hours with special care. Avoid excessive activities with birds during periods of peak heat periods (Andersson *et al.*, 2001).

Conclusion

Temperatures higher than ambient temperature may cause harmful effects on the performance, physiology, metabolism and health of laying chickens which results in huge economic losses in poultry production, particularly in tropical and arid regions throughout the globe. Since heat stress is important contributor to the poor health of poultry birds, hence an important cause of economic losses. Numerous research experiments have explored the effects of heat stress conditions on the welfare and production performance of laying hens. The detrimental impact of high ambient temperature on layer birds include reduced production, reduced growth rates, decreased appetites, decline in feed intake and laying and impaired meat/egg qualities. Recent researches have focused on the negative influences of heat stress on bird's physiology, behavior, performance, welfare and reproduction. The strategies for ameliorating heat stress in laying hens including nutritional manipulations and good husbandry/management practice

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