## The reasons for the deterioration of chicken eggshell quality at high temperatures: a review

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**Abstract.** In hot-climate regions high ambient temperature is one of the main problems of poultry farming. It is a reason for large financial losses caused by a significant decrease in the livability and productivity of poultry and the quality of products. Poor shell quality results in increased egg breakage and cracking. Such eggs lose the abilities for long-term storage or incubation and their market price is become significantly (1.5-3-fold) reduced. In the review presented the biological role of the eggshell, certain aspects of its formation, and the main reasons for the deterioration of eggshell quality in high ambient temperatures are discussed. It was shown that the eggshell quality depends on the genotype, age, feeding, health status, management conditions, etc. High ambient temperatures (above 32-35 °C) disrupt the process of eggshell formation and leads to a significant decrease in its weight, thickness, and strength. The negative impact of high ambient temperature on eggshell quality is associated with a complex set of problems, including low feed intake by hens, acid-base and mineral imbalances, physiological disorders in the endocrine system and other organs and mechanisms involved in the process of eggshell formation. The understanding of these reasons gives an opportunity for the development and implementation of targeted interventions and enables the minimization of negative impact of heat stress on eggshell quality and the efficiency of the commercial egg production.

## 1 Introduction

High ambient temperature is one of the main environmental problems of poultry farming in regions with hot climates. It annually leads to large financial losses, mainly caused by a significant decrease in the livability and productivity of poultry, the quality of products [1-6]. The degree of negative impact depends on the strength and duration of exposure to high temperatures, the age of the hens, the type and level of their productivity, veterinary well-being, the quality of feeding, a complex of technological and genetic factors, etc. [2, 7].

The shell is the main protective coating of an egg, the main functions of which being mineral nutrition and gas exchange in the embryo, resistance to mechanical and microbial impacts on the egg to preserve its integrity [8, 9]. Poor eggshell quality results in increased egg breakage and cracking, which causes significant economic damage to farms. Eggs with damaged shells lose their abilities for long-term storage or incubation, and their market price is significantly (1.5-3-fold) reduced [10].

The main indicator of the eggshell quality is its strength, which depends on the weight, thickness, chemical composition, and uniformity of distribution of the structural material [11, 12].

This work aims to summarize and analyze the currently available scientific knowledge on the reasons for the deterioration of chicken eggshell quality under high-temperature conditions, which could form a conceptual basis for our understanding of the mechanisms associated with this phenomenon.

*Some aspects of eggshell formation*. The eggshell formation is a complex process and depends on hen genotype, age, feeding, health status, management, etc. [10, 13]. High eggshell quality can be achieved only under optimal management conditions, the presence and harmonious interaction of all factors influencing this process [14].

The ability of chickens to form good quality shells largely depends on the presence of calcium in the diet and the bone tissue pool [15]. Calcium is the most important feed factor affecting shell quality. The significant need for calcium is explained by the fact that eggshells contain up to 95-96% of calcium carbonate, the rest is the organic matrix (2-3.5%), magnesium, phosphorus, and some micronutrients [16, 17]. One eggshell contains about 2.2-3 g of pure calcium [18].

Under optimal conditions of feeding and management of highly productive laying hens three sources of ionic calcium are involved in shell calcification: diet, temporary skeletal pool (medullary

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tissue), and true skeleton. Normally for the synthesis of an eggshell the share of dietary calcium is about 68%, the share of medullary tissue is 29%, and diffuse calcium extracted from the surface of the bones accounts for about 3% [19, 20].

The dietary calcium is used for the shell formation according to the simple physical laws associated with the permeability of the intestinal walls. The deposited calcium is mobilizing via the interaction of hormones with the liver (vitellogenin) and medullary structures in the bones (calcium). The formation of carbonic acid ions responds to the presence of the carbonic anhydrase enzyme in poultry. The synthesis and metabolic activity of this enzyme is controlled by estrogens and progesterone [21, 22]. It is assumed that estrogen affects the transport of Ca<sup>2+</sup> in the duodenum by increasing the regulation of calcium channels [23], but also by stimulating the conversion of vitamin D<sub>3</sub> into its biologically active form calcitriol [24].

Under optimal conditions of management and nutrition during the shell formation the blood flow in the shell gland (uterus) of the oviduct increases 4-5-fold [25]. Calcium absorption occurs mainly in the upper part of the small intestine (in the duodenum), in two ways - as a result of active transfer of calcium through the intestinal wall and with the help of a specific calcium-binding protein. Both processes are activated by vitamin  $D_3$  [21, 22].

In the stomach, under the influence of gastric juice (hydrochloric acid), most of the dietary calcium is converted into calcium chloride (CaCl<sub>2</sub>), which almost immediately dissociates into ions (Ca<sup>2+</sup>). Entering the small intestine, calcium ions react with specific calciumbinding proteins, in particular with calbindin-D28k. The main role of calbindin-D28k is the transfer of ionized calcium from the intestine to the shell gland of the oviduct [26-28]. At a "thermoneutral" temperature (20–26 °C), the degree of expression of calbindin-D28k in the intestinal segments of laying hens has the following sequence: duodenum> jejunum> ileum> cecum> large intestine [28, 29].

The concentration of calbindin-D28k in the intestinal tissues and the oviductal gland and the Ca<sup>2+</sup> transport are closely correlated [27-30]. Calbindin is found in the intestine before sexual maturity, and noticeably increases by the onset of lay [31]. In the shell gland of the oviduct calbindin appears during the formation of the shell of the first egg and disappears within 3 days after the cease of lay [30, 31]. Its concentration is proportional to the rate of Ca<sup>2+</sup> deposition in the eggshell [30, 32]. In addition to the proposed role of calbindin in Ca<sup>2+</sup> transport it can also be involved in the protection of cells from high Ca<sup>2+</sup> concentrations or from cell degradation by apoptosis, and also act as a buffer [33].

**Impact of heat stress on the eggshell quality.** High ambient temperature or so-called heat stress (above 32-35 °C) disrupts the formation of eggshell and leads to a significant decreases in the absolute and relative weight [28], thickness, and strength of the eggshell [34]. The shell quality deteriorates due to the violation of the microstructure of both its organic and mineral parts [35].

Under heat stress peripheral vessels reflexively expand. The blood flow to the skin and other external integuments increases with a simultaneous decrease in blood supply to internal organs, including intestine, ovary, and oviduct, which is a compensatory physiological response that helps the body of a hen to cope with the excessive external heat load [36].

In addition, to restore the heat balance with the environment the bird spreads its wings, reduces activity, while its respiration depth decreases approximately 5–6-fold, while the respiration rate increases [2, 37].

Frequent breathing helps the bird to reduce body temperature by evaporative cooling, but at the same time it increases the excretion of carbon dioxide (CO<sub>2</sub>), which is necessary for the formation of calcium carbonate, which is a component of the eggshell [38]. As a result, blood pH increases, and the partial pressure of carbon dioxide in arterial blood (pCO<sub>2</sub>) decreases, which in turn causes a disturbance in the circulatory acid-base balance with its shift to the alkaline side, namely, respiratory alkalosis [39]. Respiratory alkalosis at high ambient temperatures is characterized by low blood of calcium, sodium bicarbonate concentrations (NaHCO<sub>3</sub>), carbonic acid (H<sub>2</sub>CO<sub>3</sub>), and bicarbonate ions  $(\text{HCO}_3^{-})$  [40, 41].

A change in the ratio of electrolytes and blood gases has a negative impact on the formation of a high-quality eggshell [6, 42]. The process is further aggravated by the fact that heat stress suppresses the activity of the thyroid gland, which prevents the formation of the active form of vitamin  $D_3$  in the kidneys [35, 43].

The formation of poor quality shells as a result of low level of Ca in plasma at high ambient temperatures can be associated with a decrease in blood flow to the egg-forming organs in hens under thermal stress [44]. Obviously, insufficient blood flow will reduce the supply of calcium, oxygen and other nutrients to the egg production organs, which are necessary for the optimal physiological functioning of these extremely important organs for the formation of a high-quality shell, resulting in negative consequences.

It is believed that any decrease in shell thickness that occurs with an increase in respiration rate is almost certainly associated with a decrease in the ability of blood to carry Ca as a result of alkalosis [45].

High temperatures also disrupt the mineral balance of birds, and its degree depends on the type of mineral and the severity of heat stress. It was found that heat stress significantly increases the excretion of certain minerals (phosphorus, potassium, sulfur, copper, zinc, molybdenum and magnesium) [46], Na<sup>+</sup> and K<sup>+</sup>, which (along with Cl<sup>-</sup>) play an important role in homeostasis of acid-alkaline balance of hens and regulation of osmotic pressure [47]. Other authors have also shown a decrease in plasma electrolytes with an increase in ambient temperature [48]. It can be assumed that the reason for the increased excretion of electrolytes is a significant increase (3-5-fold) in water consumption during heat stress.

Some mechanisms that can explain the deterioration of eggshell quality in laying hens under heat stress are discussed in literature. One of them is a decrease in the activity of the carbonic anhydrase, which catalyzes the formation of bicarbonate ions from carbon dioxide and water, which are converted in the shell gland into eggshell's calcium carbonate [49].

The deterioration of the chicken eggshell quality at high temperatures is partly explained by a decrease in feed intake [43, 50, 51], and at the same time calcium [52] by 25-30%, as well as by an increase in water consumption and accelerated passage of feed through the gastrointestinal tract [43].

Under the influence of heat stress against the background of a decrease in feed consumption, the weight of the small intestine decreases by 22–23% and the absorbing surface of the intesstinal villi by 19% [53, 54]. High temperature leads to imbalance between the production of reactive oxygen species (ROS) and the antioxidant defense system, which results in the damages to the epithelium and to inflammatory reaction of the intestinal mucosa [55]. Other changes which can occur in the gastrointestinal tract include the decreases in the acidity and bactericidal properties of gastric juice and the shift of the balance of beneficial and harmful intestinal microflora towards an increased content of pathogenic microbial species [56, 57].

It should also be noted that under high temperature conditions the localization of the calcium-binding protein calbindin-D28k is significantly reduced in the ileum, cecum, colon, and in the shell gland (uterus) of the oviduct. It is probably partly associated with a decrease in the secretion of estrogens [28], which, as suggested by some authors [58], are involved in the induction of the synthesis of calbindin-D28k.

In addition, low calcium intake during heat stress causes bone resorption and hyperphosphatemia. An increase in the content of phosphorus in the blood suppresses the formation of calcium carbonate (CaCO<sub>3</sub>) in the shell gland of the chicken oviduct due to the outflow of blood to the skin to cool the body, thereby reducing blood flow to the uterus and its "calcium nutrition"; as a result, the eggshell quality deteriorates [35, 50]. Given that most of the calcium for the formation of eggshells comes from the diet, it can be concluded that any factor (for example, high ambient temperature) that causes a decrease in feed intake will simultaneously lead to a decrease in the level of calcium intake, its mobilization and availability, and hence directly contribute to a decrease in eggshell quality.

## 2 Conclusion

Analysis of numerous available studies shows that the negative impact of high ambient temperature on the eggshell quality is associated with a complex set of problems. These problems include low feed intake, acidbase and mineral imbalances in the body, physiological disorders in the functioning of the endocrine system and other organs and mechanisms involved in the eggshell formation process. The understanding of these reasons gives an opportunity for the development and implementation of targeted interventions. This will enable to minimize the negative impact of heat stress on eggshell quality and the efficiency of the commercial egg production.

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