Review

Embryonic development and the physiological factors that coordinate hatching in domestic chickens

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ABSTRACT Embryonic growth and development is influenced by both endogenous and exogenous factors. The purpose of this review is to discuss the critical stages of chick embryonic development in relation to functional maturation of numerous organ systems, the acquisition of thermoregulation, and the hatching process. In addition, the mechanism of hatching, including sound synchronization and hormonal and environmental stimulation, will be discussed. Finally, the importance of effective hatching synchronization mechanisms will also be highlighted.

Key words: chicken incubation, embryonic development, hatching process, synchronization, sound stimulation

INTRODUCTION

Hamburger and Hamilton (1992) described a series of stages encompassing the entire period of chicken incubation (21 d) based on the external features. They defined 3 main phases: early, middle, and late stages. The formation of organs and systems occur during the first 2 stages, followed by the growth of organs and maturation of systems during the late stage. Knowledge of the normal stages in chicken embryonic development gives a deeper understanding of the developmental process and how embryonic growth and the hatching process can be influenced and manipulated by both endogenous and exogenous factors. Although all vertebrate embryos are influenced by external environmental conditions, this is particularly true of embryos that develop outside of the mother, as occurs in avian species. In avian systems, the environment is tightly regulated by the parent(s) through the supply of nutrition to the egg and through manipulation of incubation conditions in the wild (Reed and Clark, 2011). In nature, the clutch hatches under the mother hen within a short hatch window (**HW**), which is defined as the time between the early-hatching and late-hatching chicks (Molenaar et al., 2010). In contrast, during artificial incubation,

maternally derived components of incubation control and sound communication are excluded. In the poultry industry, the HW can be as long as 48 h, in part due to a difference in genetics and handling (i.e., storage conditions) among the batches of eggs. As spread of hatch increases, the time of first access to feed and water also increases. A delay in access to feed for 1-d-old chicks impairs posthatch growth (Careghi et al., 2005; van de Ven et al., 2009). Therefore, a short HW is one way to improve animal welfare and achieve a more uniform flock.

The ability of the embryo to respond to the environment during incubation and adjust its development is fundamental. The duration of incubation and time of hatching are mainly regulated by embryonic physiology and behavior. Asynchronous hatching in clutches of bird eggs is an environmentally dependent behavior and an adaptive phenomenon among siblings (Rumpf and Tzschentke, 2010). Synchronization of hatching can occur through coordinated vocalization and is also affected by physiological parameters such as plasma thyroid and corticosterone (**CORT**) levels. The actual level of physiological parameters that occur during the hatching process may predetermine the hatching time. During the hatch, hormone systems act as critical endogenous effectors that transmit environmental information to the cells, resulting in regulation of gene expression, metabolism, and behavior (McNabb, 2007).

The purpose of this review is to highlight the timing of onset of embryo functional systems: consciousness,
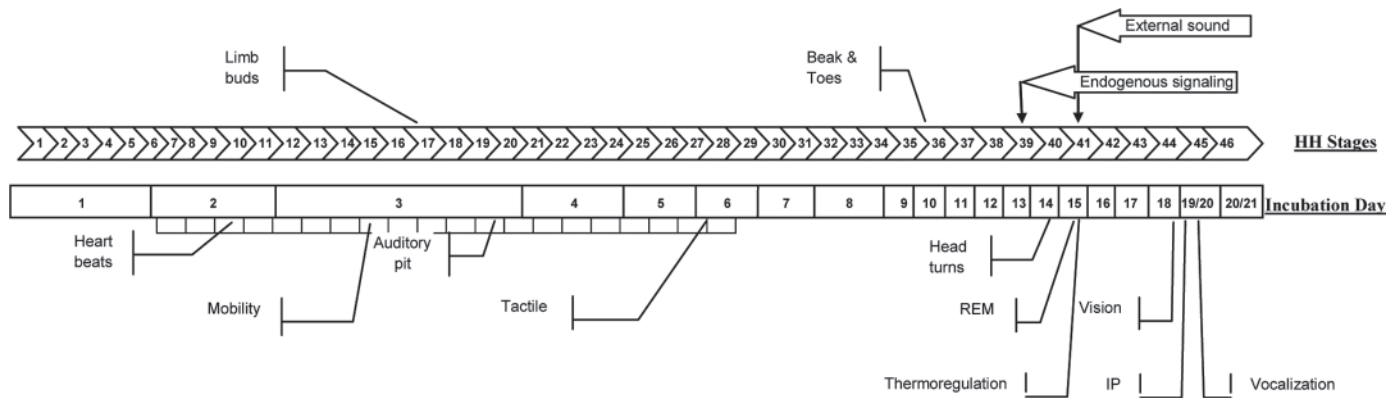
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Figure 1. Timeline and onset of major functional milestones in embryonic development. HH = Hamburger and Hamilton; REM: rapid-eye-movement; IP: internal pipping.

mobility, thermoregulation, auditory, and vocalization based on normal growth and development of the chick embryo throughout its incubation period (Figure 1). We also discuss, in detail, some mechanisms and behaviors related to the hatching process to understand effective ways to synchronize hatching. We provide evidence to suggest that the optimization of incubation conditions by manipulating the external environment can lead to a possible narrower HW.

STAGING EMBRYOS

In "A Series of Normal Stages in the Development of the Chick Embryo" (1951), Hamburger and Hamilton developed and defined specific stages of morphogenetic progression from the first cleavage divisions through hatching. The classical Hamburger and Hamilton (**HH**) Staging Table is widely used in classifying development of the chick embryo into 45 stages (Hamburger and Hamilton, 1992). In their scheme, Hamburger and Hamilton used different prominent morphological changes and developmental features to classify different phases of development. The initial stages (1 to 6) are characterized by the development of the primitive streak. The middle stages (7 to 14) are defined primarily by the number of somites and related features. The late developmental stages (15 to 45) are identified by several typical morphological features and grouped by a series of standard measurable features. The morphological changes of limbs and visceral arches are used to determine stages 18 to 40. However, toward the end of development, the chick does not undergo further morphological changes and only increases in size; therefore, these late stages require identification by objective measurements (such as the length of beak and third toe).

Eyal-Giladi and Kochav (1976) provided the first stage-by-stage procedure to describe the morphogenetic development of the early chicken embryo during preoviposition. In the Eyal-Giladi and Kochav 14-stage procedure (Eyal-Giladi and Kochav, 1976), a stage I embryo

is a blastodisc with less than 5 or so cleavage divisions and a stage XIV embryo has a completed hypoblast that is generally equivalent to stage 1 in the HH Staging Table. Thus, combining these 2 staging schemes gives a complete and comprehensive staging reference throughout chick development.

Although we recognize that the temporal rate of development differs among different species, strains, and even individuals, the external features of any embryo within any given stage will be almost identical (Sellier et al., 2006). With the detailed descriptions of a normal table as a reference, it is possible to identify the actual stage at which prenatal activation of functional systems occurs and the proper time of embryonic response. This knowledge can be applied not only to scientific research but also in the poultry industry for evaluating the effect of preincubation handling on embryo development; estimation of embryonic mortality at different stages when performing fresh egg and candling breakouts; defining normal and abnormal embryo development; and evaluating the impact of other factors (hen age, strain, and shell quality) on fertility and hatchability. These reference tables ensure that accurate, consistent, and repeatable data are obtained among laboratories and represent a standard for industry quality assessment.

DEVELOPMENT OF EMBRYO FUNCTIONAL SYSTEM

The development of organs start early in chick embryonic development, and all functional organs are well developed before hatch. Initial development of the chick embryo is rapid: by h 44 of incubation, the heart is beating and vascular systems are linked. By the end of the third day of incubation, limb buds for the wings and legs are visible and the auditory pit is established. Formation of reproductive organs, differentiation of sex, and beginning of voluntary movements take place on d 5 and 6. By d 7, the heart is completely enclosed in the thoracic cavity and feather buds are forming. By d 10, the beak hardens and toes are fully formed (stage

36 of HH when the length of beak and third toe are started to use as an indicator for embryonic staging). On embryonic d 14, the embryo is moving into position for hatching and turns its head toward the large end of egg. Rapid-eye-movement (**REM**) is detected at d 15, which is related to high levels of forebrain activation in the chick embryo and signals the time the embryo enters a growth phase before hatching (Mellor and Diesch, 2007; Mellor, 2010). Furthermore, it is shown that chicken maternal calls at this stage can stimulate the embryonic brain to a waking-like state in advance of hatch (Balaban et al., 2012).

Development of Mobility and Nervous Systems

Development of the central nervous system (**CNS**) in the chick embryo begins on the second day of incubation and matures before hatching on d 21. Embryonic movement represents the development and interaction of skeletal, muscular, and nervous systems, and these commence on d 3 of development. Embryonic limb movement serves an essential role in joint cavity formation and cartilage and bone formation during skeletal development. Restricted early embryonic movement can result in positional deformity, a reduced heart rate, reduced blood flow, oxygen and nutrient delivery, and therefore decrease growth (Lamb et al., 2003; Pitsillides, 2006). The onset sequence of activity in neurological structures underlying sensory mechanisms is: tactile by d 6, proprioceptive-vestibular by d 8 to10, taste possibly around d 12, auditory by d 12 to14, vision by d 18, and olfactory by d 20 (Rogers, 1995). With the peripheral, visceral, spinal, and brain nerve tracts developing, growing, and maturing, motor behaviors begin as spontaneous movements at d 3 (controlled individual limb, neck, and head movements) to the highly coordinated movements that prepare the body to the right position for hatching (Wu et al., 2001).

Heat Production

Energy changes and balance within an avian egg are represented by embryonic heat production (**HP**) which affects egg shell temperature. Body temperature is primarily controlled by the rate of HP and exchange with the environment. Mechanisms of HP, heat conservation, and heat loss are controlled by the central nervous system. Thermoregulation as an open loop system and a closed feedback control system occurs at d 15 and 20, respectively, when the embryo can react to the changes of incubation temperature (Tzschentke, 2008).

Different developmental stages show different HP rates during incubation (Table 1). The developmental stage-related HP of the chicken embryo gradually increases from d 9 to 16 and then reaches a plateau phase from d 17 to 19 (Janke et al., 2004). Embryonic growth rate declines shortly before hatching so that the meta-bolic rate usually plateaus before hatching in precocial birds (domestic chicken; Vleck and Vleck, 1987). At the end of the plateau phase the embryo penetrates the chorioallantoic and inner shell membranes and begins pulmonary respiration to supply the growing embryo with sufficient oxygen. The HP increases continuously from internal pipping until hatch (Wu et al., 2001; Tzschentke, 2008). Qualitatively similar developmental patterns of HP were found in all precocial and altricial bird species investigated (Tzschentke and Rumpf, 2011). Embryonic body temperature measured in the allantoic fluid also follows the same developmental pattern of HP (Janke et al., 2002). There are 2 possible explanations for this phenomenon. One is that HP is calculated by O_2 consumption and CO_2 production under constant incubation temperature; therefore, increasing metabolic rate and oxygen consumption may cause an increase in body temperature. The second is the possibility that increasing body temperature is due to an increased thermoregulatory set-point and this in turn leads to an increased HP. However, at the late stage of incubation, lung respiration (a mechanism of heat loss) is considered to be more effective in controlling body temperature than HP mechanisms via adjusting the respiration rate and tidal volume (Tzschentke, 2008). Different strains also show different HP rates (Table 1). The HP has a tendency to be higher in layers than in broilers, possibly because protein synthesis (which requires energy) is faster in layers than in broilers (Romijn and Lokhorst, 1960; Muramatsu et al., 1987; Muramatsu et al., 1990; Sato et al., 2006). Differences in HP have also been noticed between different strains of broilers (Tona et al., 2004; De Smit et al., 2005).

Auditory Development

Several studies report that the auditory development in birds is precocious during incubation (Konishi, 1973; Friauf and Lohmann, 1999; Rubel and Fritzsch, 2002). However, it is not clear when the embryo is able to detect and encode natural ambient sound, and what are the ranges of characteristic frequencies (**CF**) and threshold levels for sensitivity. The cytological differentiation of hair cells in the domestic chick occurs between d 8 and 10, and by d 10 the middle ear apparatus is also well-formed (Whitehead and Morest, 1985a,b). Synaptic contact in the hair cells of the chicken basilar papilla appear on d 10 (Rebillard and Pujol, 1983; Whitehead and Morest, 1985a,b), and central synapses between primary afferents and second-order auditory neurons in nucleus magnocellularis are functional around d 12 (Jackson et al., 1982; Pettigrew et al., 1988). The response of cochlear ganglion neurons to sound are tested, and results indicate that in domestic chickens (*Gallus gallus* L. f. *domestica*) the ontogeny of hearing begins with endogenous rhythms around the period of peripheral synaptogenesis between d 13 and 14 (HH stages 39 to 40). Cochlear primary afferent

Table 1. Mean values of heat production and body temperature during incubation in Ross 308, Ross 508, and White Leghorn chickens (Janke et al., 2004)

Incubation day	Heat production (W·kg^{-1})			Body temperature (°C)		
	Ross 308	Ross 508	White Leghorn	Ross 308	Ross 508	White Leghorn
9	0.19	0.23		37.5	37.6	
10	0.28	0.26		37.6	37.6	
11	0.33	0.46	0.44	37.6	37.8	37.9
12	0.90[a]	0.87[a]	0.57[b]	38.2[a]	38.2[a]	37.9[b]
13	1.24[a]	1.35[a]	0.82[b]	38.6[a]	38.6[a]	38.2[b]
14	1.54[a]	1.81[a]	1.17[b]	38.9[a]	39.0[a]	38.3[b]
15	2.09[a]	2.23[a]	1.47[b]	39.4[a]	39.3[a]	38.7[b]
16	2.15	2.57	1.89	39.4[ab]	39.6[a]	39.0[b]
17	2.52	2.67	2.22	39.6	39.6	39.3
18	2.61	2.49	2.17	39.7[a]	39.6[a]	39.3[b]
19	2.74[a]	2.68[a]	2.12[b]	39.7[a]	39.6[a]	39.2[b]
20	4.20[a]	3.99[a]	2.18[b]	40.5[a]	40.5[a]	39.0[b]
21			3.25			40.0

[a,b]Means within a row with different superscripts differ ($P < 0.05$).

neurons cannot encode frequency information from the natural sound environment until the sensitivity occurs and matures at stage 42. From d 15 to 16 (stages 41 to 42), cells responded to external sound at a relatively high threshold (86.0 ± 10.9 dB) and wide range of frequencies (50 to 5,000 Hz). The CF threshold levels for airborne sound stimulation decrease in older embryos (stages 43 to 44) to 60.5 ± 18.2 dB sound pressure level (which is a logarithmic measure of the effective sound pressure of a sound relative to a reference value) and CF became restricted to frequencies ≤ 2,300 Hz. Airborne CF threshold levels decreasing with embryonic stage indicate that sound transfer improves during embryonic development (Jones et al., 2006).

Previous studies show morphological and biochemical alterations in brainstem auditory nuclei and hippocampus of domestic chicks given early prenatal sound stimulation by species-specific sound at 65 dB from d 10 until hatch (Chaudhury et al., 2006, 2009; Chaudhury and Wadhwa, 2009). Increased dendritic arborization and probably enhanced synaptogenesis could be indirectly attributed to consolidation and maintenance of synapses consequent to adherent activity induced by prenatal sound stimulation (Alladi et al., 2002). These effects indicate that prenatal auditory stimulation played an important role in chick learning and memory (Kauser et al., 2011). It is possible that prenatal auditory stimulation may also play a role in the synchronization of hatching (Veterany et al., 1999b; Vergne and Mathevon, 2008).

MECHANISM OF HATCHING

Hatching Behavior

Hatching behavior is well described in literature and begins on d 16 or 17 of incubation in the chick embryo (Oppenheim, 1970, 1972; Provine, 1971, 1972; Oppenheim et al., 1978; Pittman et al., 1978). The embryo turns its body to the correct position: a folded neck and head under the air space membrane on d 18. Such neck folding is maintained after both internal and external pipping and until the chick escapes from the shell once hatching is complete. From d 19 and 20, hatching behavior is more active and vigorous head movements cause its beak to penetrate the membrane (internal pipping). The allantois, which previously served as its mode of gas exchange, begins to dry up as the chick starts pulmonary respiration. A period of relative inactivity follows, punctuated by occasional vigorous whole-body and head movements about 24 h before hatching. Then, the chick continues to push its head outward using the sharp horny structure on the upper beak (egg tooth) and the hatching muscle on the back of the neck to open a spot on the eggshell (external pipping). This is followed by another quiescent period until the hatching behavior on d 20 or 21. A series of back thrusts with the beak and egg tooth against the shell enlarges the hole made by external pipping, which, together with repeated partial body rotations achieved by stepping movements of the feet against the shell, lead to the top of the shell being loosened sufficiently to be pushed off. However, it is not clear what causes the sudden appearance of the required active behaviors after the period of inactivity that follows external pipping. External influences such as vocal-auditory interactions between the hen and chick, between chicks in the same clutch, or both, may influence the timing. Another assumption is that hatching initiates once the partial pressure of CO_2 (**pCO_2**) threshold in the air cell is reached and increases metabolic demand. Blood pCO_2 and air cell pCO_2 are increasing with embryonic development. From d 14 to 18, air cell pCO_2 increases from 15 to 34 mmHg (Bruggeman et al., 2007). Furthermore, high ambient CO_2 can cause higher blood and air cell pCO_2 in the early stage of incubation (De Smit et al., 2006; Bruggeman et al., 2007). Up to d 19, oxygen supply occurs only via blood circulating through the chorioallantoic membrane (Mortola, 2009). From d 19 to 20, however, the chick pierces the air-space membrane of the egg

with its beak (internal pipping), which allows pulmonary respiration to begin. Thereafter, until hatching is complete at about d 20 or 21, oxygen supply occurs via both the chorioallantoic membrane and the lungs. The oxygen contributed via the chorioallantois declines to low levels during this period, but the pulmonary contribution compensates with a 1.5- to 2-fold rise. This is aided at approximately d 19 or 20 by the chick penetrating the eggshell with its beak (external pipping) and thereby gaining access to atmospheric air. There needs to be an overall balance between delivery of oxygen by these means and an increase in oxygen demand by the chick as it grows, as well as the marked increase in demand associated with prehatching and hatching behavior (Menna and Mortola, 2002; Mellor and Diesch, 2007).

Vocalization and Hatching Synchronization

Hearing sensitivity, sound production, and transmission between eggs and maternal birds are necessary for the acoustic communication at late stage of embryonic development. Bird's embryos are able to produce and perceive sounds before hatching, which plays a vital role in synchronize hatching. The avian embryo produces the first calls before internal pipping, and the real vocalization gradually develops a species-specific sound after penetration of the inner egg membrane (Rumpf and Tzschentke, 2010). Then the embryos begin to regularly produce clicking sounds due to the egg tooth tapping against the eggshell. Clicks are accompanied by the development of breathing and respiration movements that are not a real vocalization but sounds controlled by the syrinx (Vince and Salter, 1967; Terskova, 1975; Hoyt and Rahn, 1980; Veterany et al., 1999b). The acoustic communication by clicking sounds is an essential factor for hatching synchronization in many bird species by either accelerating or decelerating the hatching time of embryo (Vince, 1966a,b, 1973, 1984; Brua, 2002).

Clicking sounds are well studied in quails and Muscovy ducks, but not in the domestic chicken. It has been reported that some sounds play a special role for acoustic communication in ducks. Responses to acoustic stimuli are increasing the rate of bill clicking, foot movements, eye opening, vocalization, or a combination of these. The best hearing frequency for clicking sound communication in the last quarter of incubation is a range of 300 to 600 Hz (Gottlieb, 1965; Heaton, 1972; Gottlieb, 1982; Gottlieb, 1983; Hochel et al., 2002). In the domestic chicken, hatching occurred earlier in chicks exposed to clicking sounds than in control chicks treated with no sound stimulation or maternal calls. These results suggest that chicks accelerate hatching in response to internally generated clicks but not to external stimuli such as a maternal call (White, 1984).

Based on prenatal clicking and breathing rates, metabolic rate may also be another mechanism by which hatching is synchronized in Muscovy duck embryos. When the second embryo within a clutch begins to click within a certain species-specific range, embryos adapt their clicking rates to those of others, which results in a common rhythm. Toward hatching, embryos click more and more regularly with an increased synchronized clicking rate (Vince, 1973; Rumpf and Tzschentke, 2010). However, the actual mechanism underlying clicking sound communication is not yet clear. It is also not clear whether an embryo can just hear its neighbor(s) or all the embryos in the clutch. One hypothesis is that most of the sounds the embryo hears within the prenatal period are bone-conducted and not air-conducted. Because the head of the embryo has contact with the eggshell at that time, bone conduction to the embryo's ear is possible. Besides clicking sounds, several other acoustic factors have been investigated in the hatching process: other calls from embryos, calls and bill clicking from the maternal hen, and music were all tested in laboratory experiments. However, these studies suggest that none of the acoustic signals mentioned improved hatching synchronization or served as a hatching releasing signal (Vince, 1969; Veterany et al., 1999a; Boonstra et al., 2010; Rumpf and Tzschentke, 2010; Reed and Clark, 2011). However, the communication between embryo and maternal parent by any other means (vocalizations, movements, thermal and chemical signals) is still very important for the time of hatching and epigenetic adaption (Toien et al., 1986; Field et al., 2007; Grans and Altimiras, 2007).

Thyroid and Adrenal Hormone Changes

The thyroid and adrenal glands are functional by the end of the first week of incubation in the chicken embryo. Thyroid hormone and glucocorticoid secretion are stimulated by the thyroid-stimulating hormone (**TSH**) and adrenocorticotropic hormone (**ACTH**), which have been reported to appear at d 4.5 and 7, respectively (De Groef et al., 2008). The peripheral glands are controlled and regulated when the hypothalamo-pituitary axis is formed around mid-incubation. Toward the end of incubation, endocrine axes undergo profound changes: not only stimulating general growth and differentiation of the chick embryo but also preparing it for its life outside the egg by regulating processes such as yolk sac retraction, the onset of lung respiration, hatching, and the initiation of endothermic responses. Thyroid hormones (**TH**) and glucocorticoid levels (**CORT**) are the most important hormones that act as stimulants for the hatching process. There are 2 types of TH responses in avian species embryos: developmental and metabolic. Thyroxine (**T4**) is a prohormone that is converted into bioactive 3,3′,5-triiodothyronine (**T3**), which circulates into cells and tissues. Plasma T3 is detectable at 9.5 d of incubation. Data from RIA measurement of T3 and T4 concentrations in chick embryo plasma show that they increase on d 15 before peaking. After in-
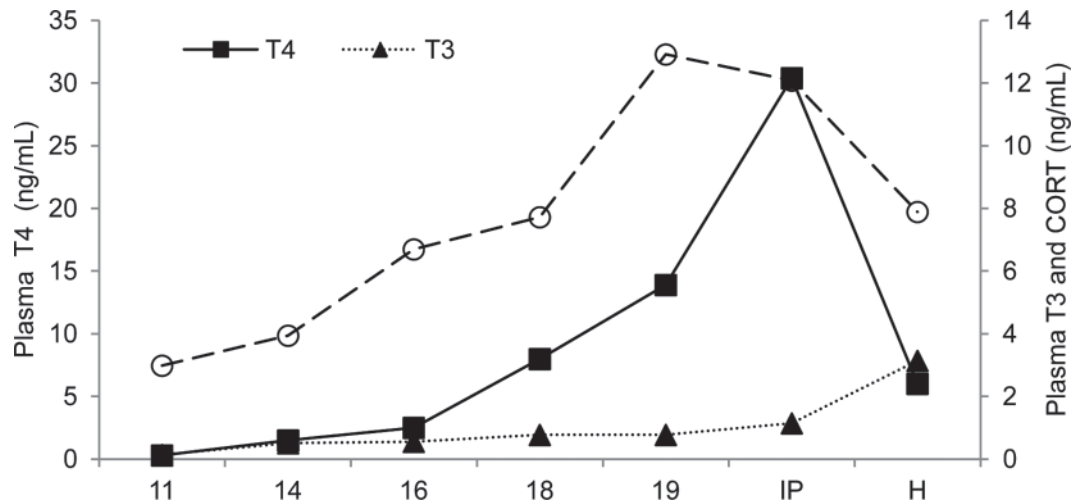
Figure 2. The 3,3′,5-triiodothyronine (T3), thyroxine (T4), and corticosterone (CORT) concentration (ng/mL) in plasma from 11-d-old embryos to newly hatched chicks (Scott et al., 1981; Reyns et al., 2003; Sechman et al., 2006). IP = internal pipping; H = hatch.

ternal pipping, T4 levels reduce, whereas T3 reaches a maximum during hatch (Figure 2). After hatch, both T4 and T3 return to low levels (Reyns et al., 2003; Lu et al., 2007). This suggests that the thyroid hormones play an important role in the hatching process between the start of pulmonary respiration and hatching. In addition, higher TH concentrations have been linked to a shorter HW. In turkey embryos, TH are being elevated to affect maturation and survival functions during the final stages of incubation, and consequently affecting hatching time (Christensen et al., 2002).

Plasma CORT is also related to embryonic development and a higher concentration leads to the initiation of chick's hatching process (Decuypere et al., 1991). The CORT can stimulate growth hormone (**GH**) secretion on embryonic d 12 in the chicken, and the secretion of CORT during embryonic development is modulated by T3 and T4 (Mashaly, 1991; Bossis and Porter, 2000). Plasma CORT concentration can be detected by d 10; then pituitary control of the adrenal function becomes important at about d 14. From d 10 to 14, the levels of CORT fluctuate slightly and then increase sharply by d 16. From this time, plasma CORT remains relatively stable through d 18 with an increasing trend until d 20 (Figure 2; Kalliecharan and Hall, 1974, 1976; Scott et al., 1981).

Prenatal stress has the potential to cause serious consequences to production agriculture and animal well-being. Therefore, absence of stress or the ability to cope with stress is a potential indicator for positive animal welfare (Archer et al., 2009). Glucocorticoids (cortisol and CORT) are stress hormones and reflect the activity of adrenal glands (Mostl and Palme, 2002).

Environmental factors can influence circulating hormone levels, thus affecting metabolic functions and the hatching process (Kuhn et al., 1984). However, the exact physiological mechanism by which this occurs is not clear. The CO_2 concentration is known to be a gaseous environmental stimulus, which could alter plasma hormones. Hypoxic conditions stimulate CORT secretion at late stages of incubation (Blacker et al., 2004). Compared with ventilated incubation (CO_2: 0.1%), chicken embryos in nonventilated incubation (CO_2 reached 1% or 1.5% at d 10) had higher plasma CORT and T3 levels at pipping, and the HW was advanced by 10 to 15 h as well as narrowed. It was demonstrated that increased CO_2 during early stages of incubation enhanced embryonic growth, chick quality, and broiler performance (De Smit et al., 2006; Bruggeman et al., 2007; Tona et al., 2007; De Smit et al., 2008). In addition, high CO_2 concentration in the air cell during the hatching phase can trigger the embryo into pipping and stimulate embryos to emerge from the egg, thereby reducing the HW (Molenaar et al., 2010). However, both the mechanism and impact on chick quality and posthatch performance is not yet clear.

CONCLUSIONS AND PERSPECTIVES

A synchronized birth is important for any species that produces many offspring in a clutch or litter as it creates an ideal environment for survival. Altering the environment of a developing organism might change the developmental trajectories of some physiological regulation systems and may alter the time of hatching and may also subsequently affect the characteristic maturity or quality of the young during the first few days' life (Tzschentke and Plagemann, 2006). For chicks, it is believed that simply reducing HW and forcing chicks to hatch without being well-developed may have a negative effect on chick quality and subsequent performance. Therefore, to achieve better chick quality and welfare, optimal incubation conditions have to be applied and the embryo response has to be considered. Besides incubation conditions (temperature, humidity, gas pressure) and embryonic physiological parameters,

other environmental factors are also important to determine the actual hatching times, such as order of ovulation, thermal microenvironments of eggs in incubation period, and intraclutch communication.

It is possible to manipulate environmental conditions to change circulating hormone levels that lead to an optimal hatchability and HW. To create an incubation system that will automatically lead to synchronized hatching, some key factors should be considered: 1) preincubation conditions; 2) incubation conditions; 3) maturity and sensitivity of embryo functional system; 4) the period and features of external stimuli; and 5) species-specific ability to synchronize. Combination of incubation conditions and microenvironment of eggs might be a way to achieve ideal flock uniformity.

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