

Hatching system and time effects on broiler physiology and posthatch growth

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ABSTRACT A multilevel housing system for broilers was developed, named Patio (Vencomatic BV, Eersel, the Netherlands), in which the hatching and brooding phase are combined. In a Patio system, climate conditions differ from those provided in the hatcheries currently in use. We compared the physiology of broilers hatched in a hatcher or in a Patio system, and included the effects of hatching time. Eggs from 1 breeder flock were incubated until embryonic d 18 in a setter and subsequently placed in a hatcher or the Patio until the end of incubation. From each hatching system, 154 chicks were collected per hatching time, at 465 h (early), 480 h (midterm), and 493 h (late) of incubation, from which 24 chicks/group were decapitated for analyses of blood plasma and organ weights. The remaining 130 chicks in each group from both systems were individually labeled and placed together in the Patio system. All chicks were given access to feed and water directly after hatch and were housed up to d 45 to monitor growth. From embryonic d 18 until the end of incubation, average ambient temperature and RH were

38.1°C and 50.8% in the hatcher and 35.2°C and 29.7% in the Patio system. Glucose and corticosterone were slightly higher in hatcher chicks, whereas organ weights were not affected by the hatching system. Although hatchling weights were lower in hatchery chicks, growth from d 0 to 45 was not affected by the hatching system. In both systems, glucose increased with hatching time, whereas lactate and triiodothyronine levels decreased. Yolk weights decreased with hatching time, whereas absolute and relative weights of the yolk-free body, intestines, stomach, lungs, and heart increased, indicating more advanced maturation of organs. Growth up to d 21 was depressed in chicks in the late group, which was possibly related to lower thyroid hormone levels at hatching. We conclude that the hatching system had minor effects on hatchling physiology and that posthatch growth and livability were not affected. Because hatching time affected broiler physiology, it seems important to take hatching time into account in future studies related to hatchling physiology.

Key words: broiler, hatching system, hatchling physiology, hatch window

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INTRODUCTION

Broiler hatching eggs are commonly incubated for 17 to 18 d in setters, after which they are candled and transferred to hatcheries for the last 3 to 4 d of incubation. Chicks hatch over a time window of approximately 24 to 48 h and are removed from the hatcheries only when the majority of the chicks have hatched (Careghi et al., 2005), leading to early posthatch periods of feed and water deprivation. Suboptimal conditions during subsequent chick handling and transport and further delays in chick placement, and thus first feed and water

intake, are associated with higher early mortality in chicks and poults (Kingston, 1979; Carver et al., 2002; Chou et al., 2004) and impaired posthatch performance (Halevy et al., 2000; Gonzales et al., 2003). The magnitude of the response to these adverse conditions seems to be influenced by the moment of hatching within the hatch window (Careghi et al., 2005) and late-hatching chicks seem to be especially vulnerable (Kingston, 1979).

An alternative system that can overcome the negative effects of early posthatch deprivation of feed and water is a system that combines the hatching process and the posthatch phase, in which feed and water are provided immediately after hatch. Such a system, named Patio (Vencomatic BV, Eersel, the Netherlands), was developed between 2002 and 2006 and has proved to function as an alternative to current hatching and brooding

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systems with regard to hatchability, early growth, and livability of broiler chicks (van de Ven et al., 2009).

In contrast to conditions typically observed in hatchers in the last phase of incubation, air temperature, RH, and air speed are lower in the Patio system, whereas the volume of air per egg is higher (van de Ven et al., 2009). In the last phase of incubation, thermal conditions can affect the development of several organs (Lekrisompong et al., 2007; Molenaar et al., 2010a), thermoregulation (Shinder et al., 2009), and muscle tissue (Piestun et al., 2008) of the chick. Consequently, differences in climate conditions between the hatcher and the Patio system may lead to a difference in posthatch physiology in the broiler, with possible long-lasting effects on posthatch performance. Effects of hatching in the Patio system on broiler physiology and posthatch growth are not known, and may vary in chicks hatching at different points within the hatch window.

We investigated the consequences of hatching in the Patio system vs. in a hatcher on organ development, plasma hormone concentrations, and growth to slaughter weight in broilers, taking into account the moment of hatching within the hatch window.

MATERIALS AND METHODS

All procedures in this study were approved by the Animal Care and Use Committee of Wageningen University, Wageningen, the Netherlands.

Incubation and Chick Management

Hatching eggs were obtained from a commercial Ross 308 breeder flock aged 47 wk. Eggs were stored for 3 d before being set in a HatchTech setter (HatchTech, Veenendaal, the Netherlands). A standard single-stage incubation program was used in the setter, with a gradually decreasing machine temperature from 38.1°C at embryonic day (E) 0 to 37.5°C at E18 of incubation. At E18, eggs were candled and apparently fertilized eggs were randomly divided. A total of 15,185 eggs were transferred to a Petersime hatcher (Petersime, Zulte, Belgium) and 16,200 eggs were transported to the Patio system (Vencomatic BV) for 30 min in a climate-controlled truck at an air temperature of 30°C.

A standard hatching program was used in the hatcher, beginning at a set point temperature of 37.2°C at E18, which was gradually decreased to 36.4°C at E21. In the Patio system, the set point of the air temperature was 35.0°C during the entire hatching process. At approximately E21.5 (after 514 h of incubation), the hatching process was ended in both hatching systems. For measurements of physiology and growth, chicks that still had some wet down, indicating they had just hatched, were randomly selected from both hatching systems. This was done at 3 points during the hatching process: after 465 h (early), 480 h (midterm), and 493 h (late) of incubation. In total, 154 chicks per hatching time per hatching system (named a hatch group) were selected,

from which 24 chicks were decapitated for blood collection and analyses of organ development. The remaining 130 chicks from each hatch group that had hatched in the hatcher were transported to the Patio system for 30 min at an air temperature of 30°C. After arrival at the Patio system, the 130 chicks from the hatchery and the 130 chicks that had hatched in the Patio system were individually marked with wing clips and placed in the Patio system, where they were given free access to water and feed. When the hatching process was ended, 390 chicks (130 early + 130 midterm + 130 late chicks) from the hatcher and 390 chicks from the Patio system were housed together, forming 1 group of 780 chicks in a separate compartment of the Patio system. Chicks were fed ad libitum with a commercially available feed and were raised under standard conditions, according to the recommendations of the breeder company, until slaughter weight was reached at d 45.

Data Collection

During the entire hatching process in both hatching systems, the temperature and RH were logged every 5 min using data loggers (175-H2 Logger, Testo, Almere, the Netherlands). In the hatcher, 3 loggers were placed among the eggs in different hatching baskets. In the Patio system, 3 loggers were placed among the eggs on the egg trays.

Chick BW were determined immediately after chick collection and at different ages posthatch: at E21.5 (hereafter termed d 0, meaning the normal day of placement in the broiler house) and on d 7, 21, and 45. Chicks were weighed between 1000 and 1600 h in random order. At the time of weighing on d 0, the mean biological age of chicks in the early group was 53 h, that of the chicks in the midterm group was 38 h, and that of the chicks in the late group was 25 h. Growth per hour was calculated for different growth periods: hatch to d 0, d 0 to 7, d 7 to 21, and d 21 to 45. During the last weighing on d 45, the sex of all birds was determined.

Blood Plasma and Organ Analysis

For blood sampling, 24 chicks/hatch group were decapitated within 1.5 h after collection from the hatching system. Until that time, none of the chicks had access to feed or water. Blood samples were collected in 4-mL fluoride tubes (BD Vacutainer, Becton Dickinson, Franklin Lakes, NJ) to which 10 μ L of heparin was added, and blood samples were placed on ice immediately after collection. When 6 samples were collected, the tubes were centrifuged for 6 min at $2,300 \times g$ at 10°C using a Rotofix 32 centrifuge (Hettich Zentrifugen, Tuttingen, Germany). Plasma was collected and stored in 0.2-mL Eppendorf tubes at -20°C until analysis.

Triiodothyronine (**T₃**) and thyroxine (**T₄**) concentrations were measured in plasma samples by RIA as described by Darras et al. (1992), using antisera and

T₃ and T₄ standards (Byk-Belga, Brussels, Belgium). Corticosterone concentrations were analyzed as described previously (Decuyper et al., 1983; Meeuwis et al., 1989), using a double-antibody RIA-kit (IDS Ltd., Boldon, UK). Plasma glucose, lactate, and uric acid were analyzed by colorimetric diagnostics using a biochemical analyzer (VetTest 8008, Idexx Laboratories Inc., Westbrook, ME).

The residual yolk, heart, liver, stomach (proventriculus plus gizzard), intestines, and lungs were dissected and weighed, and the relative organ weight was calculated as the ratio of organ weight to BW. The sex of each chick was determined by visual inspection after dissection.

Statistical Analyses

All data were analyzed with the SAS 9.1 software package (SAS Institute, 2004). Individual chick measurements were treated as the experimental unit in all statistical analyses. Nonnormally distributed data were log-transformed before analyses. Data on plasma hormone concentrations, absolute and relative organ weights, chick BW, and growth per hour were analyzed using the GLM procedure of SAS, according to the following model: $Y_{ijkl} = \text{sex}_i + \text{HT}_j + \text{SYS}_k + \text{interaction terms} + e_{ijkl}$, where sex_i is the sex of the chick, HT_j is the hatching time (early, midterm, late), SYS_k is the hatching system (hatchery, Patio), and e_{ijkl} is the residual error term. Data on mortality and sex distribution were analyzed using the LOGISTIC procedure of SAS, using the following model: $Y_{ijk} = \text{HT}_i + \text{SYS}_j + \text{interaction term} + e_{ijk}$.

In all analyses, P -values ≤ 0.05 were considered statistically significant and nonsignificant interaction terms were deleted from the model. When the means of the GLM were statistically different, means were compared using least squares means with Tukey's adjustment for multiple comparisons. Data are expressed as least squares means \pm SE unless otherwise stated.

RESULTS

Registrations of temperature and RH in both hatching systems were used from 449 h of incubation until the end of incubation. The mean \pm SE, minimum, and maximum temperatures were 38.1 ± 0.02 , 36.3°C , and 40.1°C in the hatcher and 35.2 ± 0.02 , 32.7 , and 36.0°C in the Patio system. The mean \pm SE, minimum, and maximum RH were 50.8 ± 0.54 , 22.9 , and 89.8% in the hatcher and 29.7 ± 0.17 , 18.0 , and 46.1% in the Patio system.

Hatching Time

Female chicks hatched earlier than male chicks ($P < 0.01$; Figure 1). Chick BW at each age are presented in Table 1. No difference in hatch weight was observed among hatching times, but BW gain per hour from

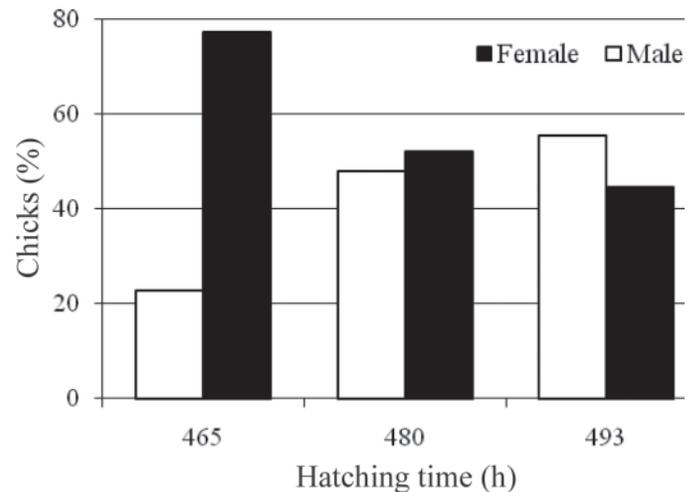


Figure 1. Sex distribution in chicks hatched early (465 h), midterm (480 h), or late (493 h) in the hatching process.

hatch to d 0 decreased with hatching time ($P < 0.01$). At d 0, chick BW were higher among chicks in the early group (52.9 ± 0.3 g), compared with those in the midterm (51.0 ± 0.3 g; $P < 0.01$) and late (48.3 ± 0.3 g; $P < 0.01$) groups. From d 0 to 7, growth per hour decreased with hatching time ($P < 0.01$; Figure 2). Day 7 BW were higher for chicks in the early group (168.4 ± 1.4 g) compared with those in the midterm (162.7 ± 1.3 g; $P < 0.01$) and late groups (151.8 ± 1.3 g; $P < 0.01$). From d 7 to 21, chicks in the late group showed lower growth than chicks in the early ($P = 0.05$) and midterm groups ($P = 0.02$). At d 21, chicks in the early (823 ± 8 g) and midterm groups (821 ± 7 g) were heavier than those in the late group (788 ± 7 g; $P < 0.01$). The effect of hatching time on chick growth was not affected by hatching system or sex. From d 21 to 45, growth was not affected by hatching time, and d 45 BW did not differ among hatching times.

Organ weights are presented in Figure 3. All absolute and relative organ weights, except yolk weights, increased from the early and midterm groups to the late group. Both absolute and relative yolk weights decreased with hatching time ($P < 0.01$). Yolk-free body mass was calculated as BW minus yolk weight, and was not affected by hatching time.

Concentrations of blood parameters at hatch are presented in Figure 4. Chicks in the late group had higher glucose than those in the early and midterm groups ($P < 0.01$). Lactate was higher in chicks in the early group than in those in the late group ($P < 0.01$), with chicks in the midterm group being intermediate and not different from the other groups. No effect of hatching time on corticosterone level was observed.

Hatching System

The sexes were distributed equally across the hatching systems. At hatch, chicks in the Patio system were heavier (48.7 ± 0.2 g) than those in the hatcher (47.4

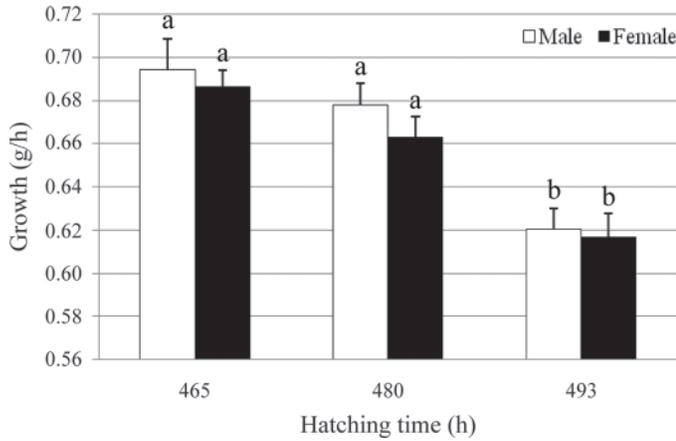


Figure 2. Growth per hour from d 0 to 7 of chicks hatched early (465 h), midterm (480 h), or late (493 h) in the hatching process in a hatcher (Petersime, Zulte, Belgium) or a Patio system (Vencomatic BV, Eersel, the Netherlands). Data lacking a common letter (a, b) are significantly different ($P < 0.05$).

± 0.2 g; $P < 0.01$). From hatch to d 0, chicks in the hatcher gained more BW per hour than chicks in the Patio system ($P < 0.01$). Although given immediate access to feed, chicks in the late group of the Patio system lost BW during the first 25 h posthatch. From d 0 onward, chick BW or growth per hour was no longer affected by hatching system.

No main effects of hatching system were observed for any of the absolute or relative organ weights (Figure 3). Plasma glucose was higher for chicks in the hatcher than for those in the Patio system ($P = 0.02$; Figure 4). For lactate level, no difference was observed between chicks in the hatcher and in the Patio system. Corticosterone was higher for chicks in the hatcher than for those in the Patio system ($P = 0.02$).

Hatching Time \times Hatching System

Overall mortality at d 45 was 2.92% ($n = 23$) and showed an interaction of hatching time \times hatching system ($P = 0.01$). For chicks in the hatcher, d 45 mortality was 0.72, 0.77, and 6.98% for chicks in the early, midterm, and late groups, respectively, and for chicks in the Patio system, d 45 mortality was 3.88, 2.96, and 2.36% for chicks in the early, midterm, and late groups, respectively. Of the 6.98% mortality ($n = 9$) for late-hatching chicks from the hatcher, approximately half ($n = 4$) died before d 7 for no clear reason.

For both absolute and relative stomach weights, a hatching time \times hatching system interaction was observed ($P < 0.01$). In chicks in the late group, stomach weights were higher for those in the Patio system than for those in the hatcher ($P < 0.01$), whereas no differences between hatching systems were observed for chicks in the early and midterm groups.

For uric acid, T_3 , and T_4 , an interaction of hatching time \times hatching system was observed ($P < 0.01$). Uric acid was higher for chicks in the late group than for

Table 1. Least squares means (\pm SE) of weights (g) of approximately 130 chicks hatched early (465 h), midterm (480 h), or late (493 h) in the hatching process in a hatcher¹ or in a Patio system²

Item	Early				Midterm				Late			
	Males		Females		Males		Females		Males		Females	
	Hatcher	Patio										
Hatch	47.0 \pm 0.6	48.0 \pm 0.7	47.5 \pm 0.4	48.4 \pm 0.4	47.4 \pm 0.5	48.6 \pm 0.5	47.8 \pm 0.4	49.1 \pm 0.4	47.6 \pm 0.4	48.7 \pm 0.5	47.0 \pm 0.5	48.9 \pm 0.5
d 0	53.3 \pm 0.8	51.5 \pm 0.9	53.5 \pm 0.5	52.5 \pm 0.5	51.1 \pm 0.6	51.5 \pm 0.6	50.8 \pm 0.6	50.6 \pm 0.6	48.1 \pm 0.5	48.4 \pm 0.6	48.1 \pm 0.7	48.3 \pm 0.6
d 7	170.4 \pm 3.6	165.9 \pm 4.2	168.9 \pm 2.0	166.5 \pm 2.2	164.3 \pm 2.7	164.8 \pm 2.7	159.8 \pm 2.5	162.3 \pm 2.5	149.9 \pm 2.4	154.7 \pm 2.6	152.5 \pm 3.0	150.8 \pm 2.8
d 21	847 \pm 18	869 \pm 21	798 \pm 10	780 \pm 11	851 \pm 13	839 \pm 13	781 \pm 12	811 \pm 12	799 \pm 12	820 \pm 13	775 \pm 15	761 \pm 14
d 45	2,772 \pm 50	2,937 \pm 57	2,462 \pm 28	2,434 \pm 30	2,805 \pm 37	2,757 \pm 37	2,452 \pm 35	2,515 \pm 35	2,731 \pm 34	2,734 \pm 37	2,467 \pm 41	2,466 \pm 39

¹Petersime hatcher (Petersime, Zulte, Belgium).

²Vencomatic BV (Eersel, the Netherlands).

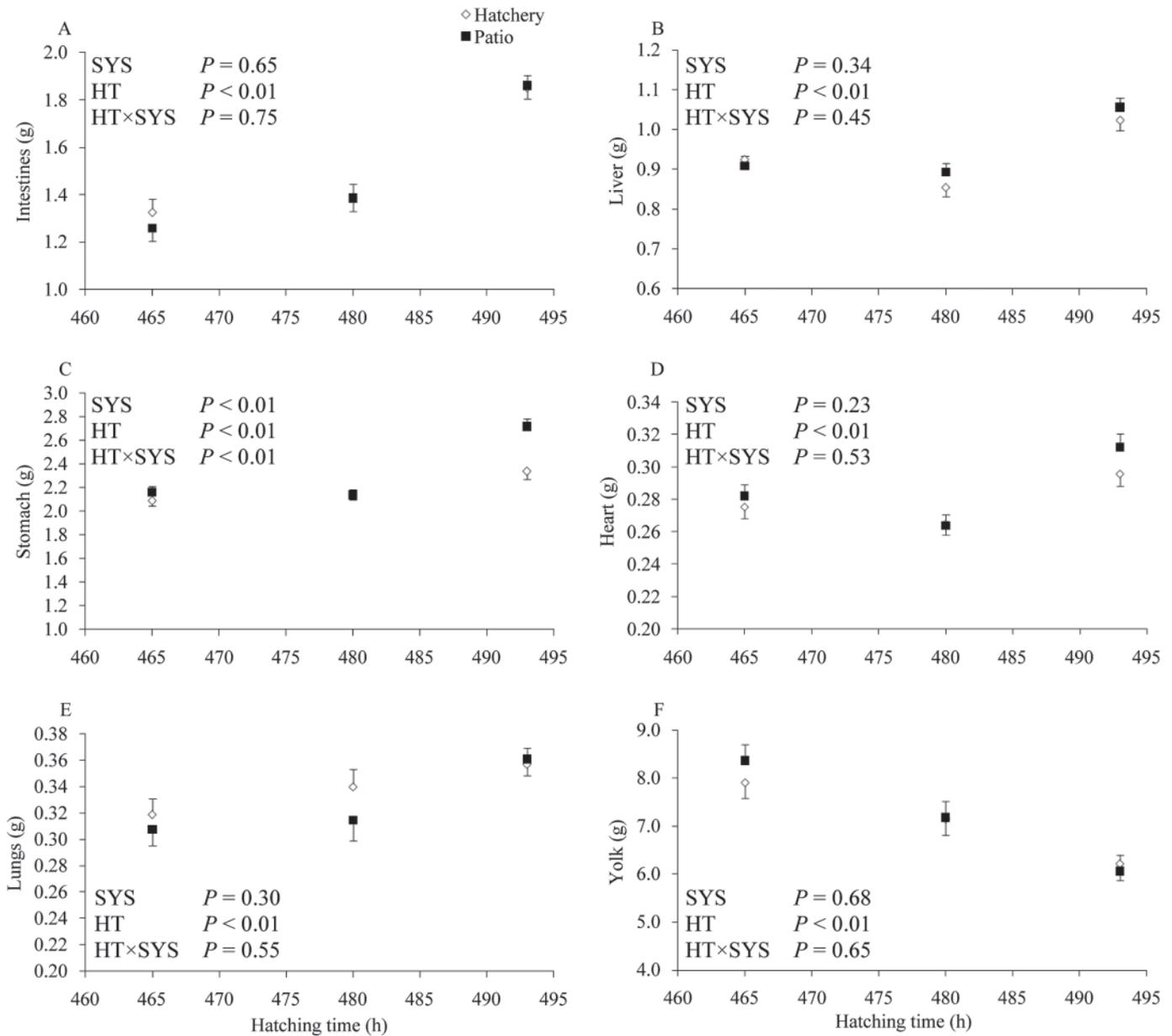


Figure 3. Least squares means of organ weights at hatch of chicks hatched early (465 h), midterm (480 h), or late (493 h) in the hatching process in a hatcher (Petersime, Zulte, Belgium) or a Patio system (Vencomatic BV, Eersel, the Netherlands). A) Intestines; B) liver; C) stomach; D) heart; E) lungs; F) yolk. SYS = hatching system; HT = hatching time; HT \times SYS = hatching time \times hatching system interaction.

those in the midterm and early groups in the hatcher ($P < 0.01$), whereas for chicks in the Patio system, no differences in uric acid level were observed among hatching times. In chicks in the early group, uric acid was higher for chicks in the Patio system than for those in the hatcher ($P < 0.01$). Plasma T_3 was higher for chicks in the hatcher than for those in the Patio system only in the midterm group ($P < 0.01$), whereas for chicks in the early and late groups, T_3 did not differ between hatching systems. A general trend was observed for decreasing T_3 with hatching time: for chicks in the hatcher, plasma T_3 was lower in chicks in the late group than in those in the early and midterm groups ($P < 0.01$), and for chicks in the Patio system, T_3 was lower for chicks in the late and midterm groups than for those

in the early group ($P < 0.01$). A comparable interaction between hatching time \times hatching system was observed for T_4 , which was higher for chicks in the hatcher than for those in the Patio system only in the midterm group ($P < 0.01$). For chicks in the Patio system, those in the midterm group had lower T_4 levels than those in the early ($P < 0.01$) and late ($P = 0.05$) groups. The $T_3:T_4$ ratio was higher for chicks in the midterm group than for those in the late group ($P < 0.01$), with chicks in the early group being intermediate (data not shown).

Sex

The sexes were not distributed equally in the experimental chicks ($P < 0.01$); 58.16% of all chicks were

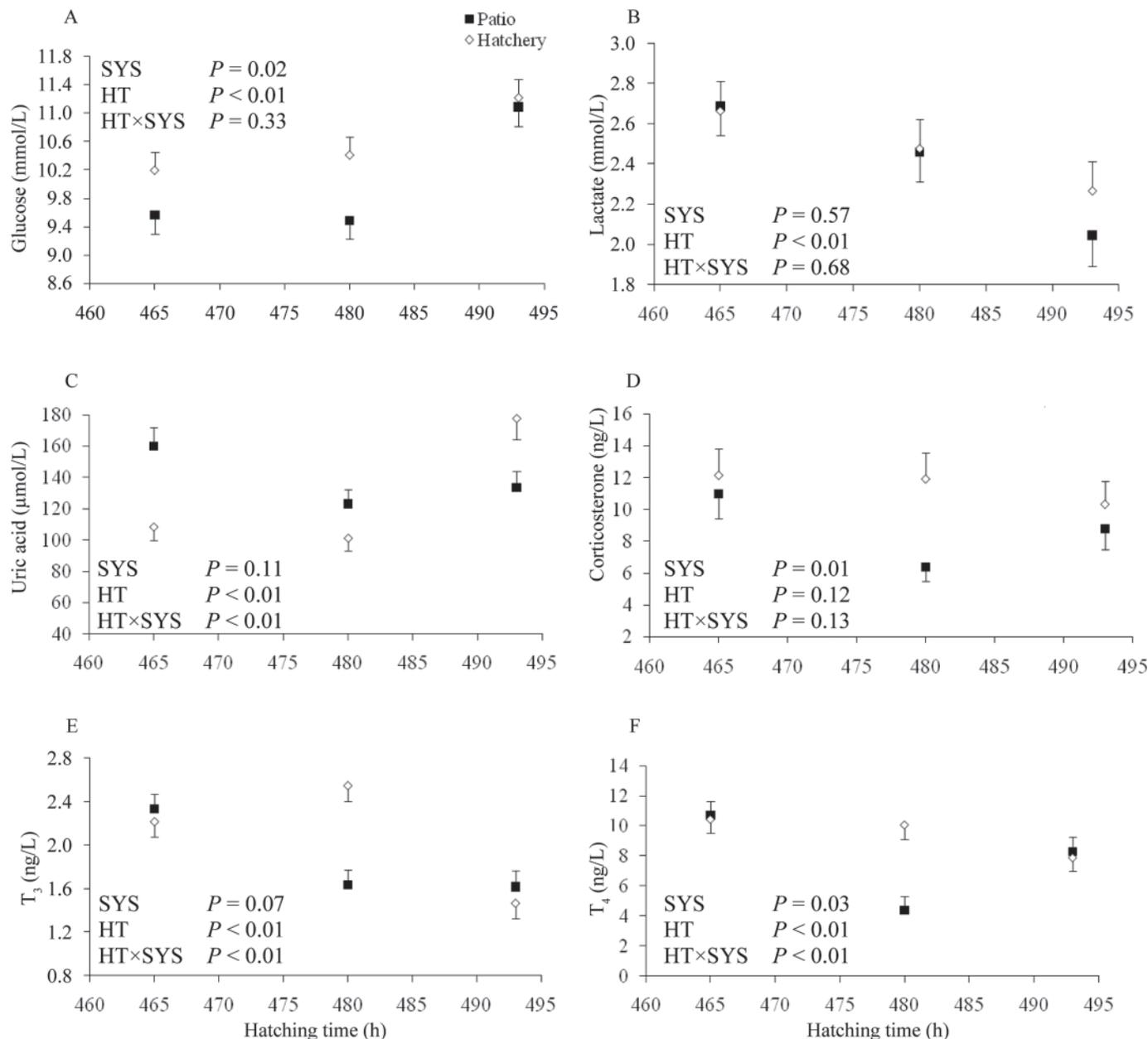


Figure 4. Least squares means of plasma hormone concentrations in chicks hatched early (465 h), midterm (480 h), or late (493 h) in the hatching process in a hatcher (Petersime, Zulte, Belgium) or a Patio system (Vencomatic BV, Eersel, the Netherlands). A) Glucose; B) lactate; C) uric acid; D) corticosterone, E) triiodothyronine (T_3), F) thyroxine (T_4). SYS = hatching system; HT = hatching time; HT \times SYS = hatching time \times hatching system interaction.

female. No effects of sex were observed on hatch weight, on growth from d 0 to 7, or on BW at d 7. From d 7 to 21 and d 21 to 45, males showed higher growth rates than females ($P < 0.01$). Males were heavier than females at d 21 (837 ± 6 vs. 784 ± 5 g, respectively; $P < 0.01$) and at d 45 ($2,786 \pm 17$ vs. $2,466 \pm 14$ g, respectively; $P < 0.01$). Overall, BW gain/h from hatch to d 45 was affected only by sex, with higher growth in males (2.45 ± 0.02 g/h) than in females (2.16 ± 0.01 g/h). No effects of sex were observed for absolute or relative organ weights or plasma hormone concentrations at hatch.

DISCUSSION

Hatching Time

Hatching time is known to be influenced by factors such as parental age, storage time and storage conditions, and incubation conditions (Tona et al., 2003; Careghi et al., 2005; Decuyper and Bruggeman, 2007). In the current study, these factors were standardized as much as possible: eggs were obtained from a single breeder flock, produced on the same day of lay, stored in one storage room, and incubated simultaneously in the

same single-stage incubator. However, there was still a spread of hatch of approximately 30 h in both hatching systems. Additional intrinsic factors influencing hatching time are the order of an egg in a clutch and egg weight (Meijer and Siemers, 1993; Careghi et al., 2005), which were not controlled in the present study.

In agreement with the findings of Careghi et al. (2005), chick BW at hatch were not affected by hatching time, but early growth was lower in chicks in the late group. In accordance with the present data, decreased chick quality and chick performance to 7 d in late-hatching chicks were linked to lower thyroid levels (Decuyper et al., 1990; Buys et al., 1998; Careghi et al., 2005; Decuyper and Bruggeman, 2005). The reason for the low quality of late-hatching chicks is not clear, but studies in other avian species have pointed to a relationship with yolk androgen levels of maternal origin and the order of an egg in a clutch (Eising et al., 2001; Müller et al., 2004). In the present study, because no data were available on yolk androgen levels or the order of an egg in a clutch, such influences could not be examined.

The present data showed increasing organ weights and decreasing yolk weights with increasing hatching time, indicating a more advanced maturation of organs with longer hatching times, as proposed by Ricklefs (1987) in a comparison among avian species. The higher organ weights in chicks in the late group in the current study coincided with lower posthatch growth, indicating the relation between organ weights at hatch and organ functionality or growth potential post hatch is weak and needs further investigation.

In the current study, plasma glucose at hatch increased and lactate decreased with hatching time. Christensen et al. (2000, 2001) demonstrated that the time of hatch, but also the duration of the hatching process itself, showed a large variation, which was associated with differences in energy availability in this phase. The energy required for hatching activities comes from glucose provided from glycogen in the liver and muscle tissues (Freeman, 1969), resulting in an increase in plasma glucose between pipping and hatch (Freeman, 1965, 1969; Christensen et al., 2001). As hatching progresses, oxygen becomes limiting and energy comes from anaerobic glycolysis, leading to increases in plasma lactate (Tazawa et al., 1983; Høiby et al., 1987; Moran, 2007). Hypoxic conditions end at the moment of external pipping, and as soon as oxygen availability is restored, birds can recycle lactate back into glucose in the liver (De Oliveira et al., 2008). The combination of high glucose and low lactate in late-hatching chicks may point to an increased interval between external pipping and hatching, enabling chicks in the late-hatching group to recycle lactate back into glucose before emergence from the egg. Alternatively, late-hatching chicks may have used less glucose and produced less lactate during the hatching process because of a lower

metabolism. In the current experiment, lower thyroid levels were found in late-hatching chicks, suggesting a lower metabolism (Decuyper et al., 1990), which could be linked to greater external pipping-to-hatching intervals (Tona et al., 2007; Everaert et al., 2008).

Hatching System

During hatching, air temperature and RH were higher in the hatcher than in the Patio system, but air speed was lower in the Patio system, which makes it difficult to quantify sensible and latent heat losses. However, hatch weights were lower for chicks in the hatcher than in the Patio system, which could point to an increased dissipation of latent heat from the embryos in the hatcher. Wineland et al. (2006) also found higher hatch weights in chicks incubated at a lower temperature (36°C) than in chicks incubated at a high (39°C) temperature from E17 to E21. From hatch to d 0, chicks in the hatcher gained more BW than chicks in the Patio system, which may be related to a greater need to compensate for the moisture loss in the last phase of incubation by a higher water intake. Chick BW at d 0 and chick growth from d 0 to 45 were not affected by the hatching system.

Despite differences in climate conditions, and in hatchling BW and early growth between the hatching systems, no clear differences were found in absolute or relative organ weights. In previous studies, high temperatures (>38.8°C) in the last week of incubation led to lower chick BW (Leksrisompong et al., 2007; Lourens et al., 2007), lower heart (Givisiez et al., 2001; Leksrisompong et al., 2007; Lourens et al., 2007; Molenaar et al., 2010a), gizzard, proventriculus, and small intestine weights, and higher yolk sac weights (Leksrisompong et al. 2007; Molenaar et al., 2010a) compared with control chicks that were incubated at 37.8 to 38.2°C. The difference between these studies and the present study can possibly be explained by the timing and duration of exposure to high temperatures. In the present study, eggs were exposed for 2 to 3 d from E18 until hatching, whereas in previous studies, exposure was from E9 to E15 onward. In addition, temperatures applied in the high-temperature groups in previous studies were higher than the mean temperature measured in the hatcher in the current study (38.1°C), which may explain the different findings. It can be speculated that from E18 to E21, embryos were able to maintain normal organ development at air temperatures in the range of 35.2 to 38.2°C, as applied in the present study.

Plasma glucose and corticosterone at hatch were slightly higher for chicks in the hatcher than for those in the Patio system. Higher corticosterone could point to a more energy-demanding hatching process (Piestun et al., 2008), resulting in increased gluconeogenesis (Joseph and Ramachandran, 1992) and higher plasma glucose at hatch.

Hatching Time × Hatching System

The interaction between hatching time and hatching system on uric acid and thyroid hormone levels suggests that although the length of the hatch window was not greatly affected, the hatching peak may have been delayed for chicks in the hatcher. High temperatures in the last incubation phase increase energy use from the anaerobic system by the embryo and suppress embryo development (Lourens et al., 2006). Under hatcher conditions, chicks in the midterm group may have hatched relatively early within the hatch window and, consequently, resemble chicks in the early group, whereas chicks in the midterm group in the Patio system may have hatched relatively late within the hatch window, and physiologically resemble chicks in the late group.

For chicks in the hatcher, uric acid was higher in chicks in the late group than for those in the midterm and early groups, whereas for chicks in the Patio system, no differences in uric acid level were observed among hatching times. Uric acid is a major nitrogenous waste product resulting from protein catabolism (Harr, 2002) and was found to increase in broilers subjected to heat stress at d 21 posthatch (Yalçın et al., 2009). It can be speculated that in the last incubation phase, prolonged exposure to a higher temperature may be related to a greater need for gluconeogenesis (Christensen et al., 2007) to fuel anaerobic metabolism by using amino acids as a substrate (Lourens et al., 2006; Molenaar et al., 2010b), resulting in higher uric acid levels for chicks in the late group compared with those in the early and midterm groups.

Sex

At hatch, no effects of sex were observed on organ weights or any of the plasma hormone concentrations, corresponding to the findings of Lu et al. (2007). In addition, no differences in hatch weight were found between sexes, which agrees with earlier literature (Burke, 1992; Reis et al., 1997; Joseph and Moran, 2005; Lekrisompong et al., 2007). Females showed earlier hatching times than males, which is consistent with previous findings in chickens (Burke, 1992; Reis et al., 1997). The reason for sex-linked differences in hatching time in chickens remains unclear. In the present study, sex affected growth from d 7 onward, and this effect became more pronounced as the chicks aged, with males showing higher growth rates than females. Higher growth rates in broiler males were linked to higher plasma growth hormone concentrations and better growth hormone receptor occupancy (Kühn et al., 1996), and are in accordance with performance expectations provided by the breeder company.

In conclusion, the present findings show a large effect of hatching time on the physiology of newly hatched broilers. Lower early growth was observed in chicks that hatched late in the hatch window compared with

chicks that hatched early or at the peak of the hatch window. This physiological variation related to age differences in one batch of day-old chicks is rarely considered, but it can be advised to take the moment of hatch into account in future studies on hatchling physiology. Although there were marked differences between climate conditions in the hatcher and the Patio system, physiological differences in chicks at hatch were limited and posthatch growth and livability were not affected.

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REFERENCES

- Burke, W. H. 1992. Gender differences in incubation length and hatching weights of broiler chicks. *Poult. Sci.* 71:1933-1938.
- Buys, N., E. Dewil, E. Gonzales, and E. Decuypere. 1998. Different CO₂ levels during incubation interact with hatching time and ascites susceptibility in two broiler lines selected for different growth rate. *Avian Pathol.* 27:605-612.
- Careghi, C., K. Tona, O. Onagbesan, J. Buyse, E. Decuypere, and V. Bruggeman. 2005. The effects of the spread of hatch and interaction with delayed feed access after hatch on broiler performance until seven days of age. *Poult. Sci.* 84:1314-1320.
- Carver, D. K., J. Fetrow, T. Gerig, K. K. Krueger, and H. J. Barnes. 2002. Hatchery and transportation factors associated with early poult mortality in commercial turkey flocks. *Poult. Sci.* 81:1818-1825.
- Chou, C. C., D. D. Jiang, and Y. P. Hung. 2004. Risk factors for cumulative mortality in broiler chicken flocks in the first week of life in Taiwan. *Br. Poult. Sci.* 45:573-577.
- Christensen, V. L., J. L. Grimes, W. E. Donaldson, and S. Lerner. 2000. Correlation of body weight with hatchling blood glucose concentration and its relationship to embryonic survival. *Poult. Sci.* 79:1817-1822.
- Christensen, V. L., M. J. Wineland, G. M. Fasenko, and W. E. Donaldson. 2001. Egg storage effects on plasma glucose and supply and demand tissue glycogen concentrations of broiler embryos. *Poult. Sci.* 80:1729-1735.
- Christensen, V. L., M. J. Wineland, J. L. Grimes, E. O. Ovido, P. S. Mozdziak, D. T. Ort, and K. M. Mann. 2007. Effect of incubator temperature and oxygen concentration at the plateau stage in oxygen consumption on turkey embryo muscle growth and development. *Int. J. Poult. Sci.* 6:406-412.
- Darras, V. M., T. J. Visser, L. R. Berghman, and E. R. Kühn. 1992. Ontogeny of type I and type III deiodinase activities in embryonic and post hatch chicks: Relationship with changes in plasma triiodothyronine and growth hormone levels. *Comp. Biochem. Physiol. A Comp. Physiol.* 103:131-136.
- De Oliveira, J. E., Z. Uni, and P. R. Ferket. 2008. Important metabolic pathways in poultry embryos prior to hatch. *World's Poult. Sci. J.* 64:488-499.

- Decuyper, E., and V. Bruggeman. 2005. Endocrine aspects of development: New challenges for the control of incubation process. *World's Poultry Sci. J.* 61:278–284.
- Decuyper, E., and V. Bruggeman. 2007. The endocrine interface of environmental and egg factors affecting chick quality. *Poult. Sci.* 86:1037–1042.
- Decuyper, E., E. Dewil, and E. R. Kühn. 1990. The hatching process and the role of hormones. Pages 239–256 in *Avian Incubation*. S. G. Tullett, ed. Butterworth-Heinemann, Oxford, UK.
- Decuyper, E., C. G. Scanes, and E. R. Kühn. 1983. Effect of glucocorticoids on circulating concentrations of thyroxine (T_4) and triiodothyronine (T_3) and on peripheral monoiodination in pre- and post hatching chickens. *Horm. Metab. Res.* 15:233–236.
- Eising, C. M., C. Eikenaar, H. Schwabl, and T. G. G. Groothuis. 2001. Maternal androgens in black-headed gull (*Larus ridibundus*) eggs: Consequences for chick development. *Proc. Biol. Sci.* 268:839–846.
- Everaert, N., H. Willemsen, L. De Smit, A. Witters, J. De Baeremaeker, E. Decuyper, and V. Bruggeman. 2008. Comparison of a modern broiler and layer strain during embryonic development and the hatching process. *Br. Poult. Sci.* 49:574–582.
- Freeman, B. M. 1965. The importance of glycogen at the termination of the embryonic existence of *Gallus domesticus*. *Comp. Biochem. Physiol.* 14:217–222.
- Freeman, B. M. 1969. The mobilization of hepatic glycogen in *Gallus domesticus* at the end of incubation. *Comp. Biochem. Physiol.* 28:1169–1176.
- Givisz, P. E. N., M. M. da Silva, C. M. Mazzi, M. I. T. Ferro, J. A. Ferro, E. Gonzales, and M. Macari. 2001. Heat or cold chronic stress affects organ weights and Hsp70 levels in chicken embryos. *Can. J. Anim. Sci.* 81:83–87.
- Gonzalez, E., N. Kondo, E. S. P. B. Saldanha, M. M. Loddy, C. Careghi, and E. Decuyper. 2003. Performance and physiological parameters of broiler chickens subjected to fasting on the neonatal period. *Poult. Sci.* 82:1250–1256.
- Halevy, O., A. Geyra, M. Barak, Z. Uni, and D. Sklan. 2000. Early post hatch starvation decreases satellite cell proliferation and skeletal muscle growth in chicks. *J. Nutr.* 130:858–864.
- Harr, K. E. 2002. Clinical chemistry of companion avian species: A review. *Vet. Clin. Pathol.* 31:140–151.
- Høiby, M., A. Aulie, and P. O. Bjønnes. 1987. Anaerobic metabolism in fowl embryos during normal incubation. *Comp. Biochem. Physiol. A Comp. Physiol.* 86:91–94.
- Joseph, J., and A. V. Ramachandran. 1992. Alterations in carbohydrate metabolism by exogenous dexamethasone and corticosterone in post hatch White Leghorn chicks. *Br. Poult. Sci.* 33:1085–1093.
- Joseph, N. S., and E. T. Moran Jr. 2005. Effect of flock age and postemergent holding in the hatcher on broiler live performance and further-processing yield. *J. Appl. Poult. Res.* 14:512–520.
- Kingston, D. J. 1979. Some hatchery factors involved in early chick mortality. *Aust. Vet. J.* 55:418–421.
- Kühn, E. R., V. M. Darras, C. Gysemans, E. Decuyper, L. R. Berghman, and J. Buyse. 1996. The use of intermittent lighting in broiler raising. 2. Effects on the somatotrophic and thyroid axes and on plasma testosterone levels. *Poult. Sci.* 75:595–600.
- Leksrisompong, N., H. Romero-Sanchez, P. W. Plumstead, K. E. Brannan, and J. Brake. 2007. Broiler incubation. 1. Effect of elevated temperature during late incubation on body weight and organs of chicks. *Poult. Sci.* 86:2685–2691.
- Lourens, A., R. Molenaar, H. van den Brand, M. J. W. Heetkamp, R. Meijerhof, and B. Kemp. 2006. Effect of egg size on heat production and the transition of energy from egg to hatchling. *Poult. Sci.* 85:770–776.
- Lourens, A., H. van den Brand, M. J. W. Heetkamp, R. Meijerhof, and B. Kemp. 2007. Effects of eggshell temperature and oxygen concentration on embryo growth and metabolism during incubation. *Poult. Sci.* 86:2194–2199.
- Lu, J. W., J. P. McMurtry, and C. N. Coon. 2007. Developmental changes of plasma insulin, glucagon, insulin-like growth factors, thyroid hormones, and glucose concentrations in chick embryos and hatched chicks. *Poult. Sci.* 86:673–683.
- Meeuwis, R., R. Michielsen, E. Decuyper, and E. R. Kühn. 1989. Thyrotropic activity of the ovine corticotropin-releasing factor in the chick embryo. *Gen. Comp. Endocrinol.* 76:357–363.
- Meijer, T., and I. Siemers. 1993. Incubation development and asynchronous hatching in junglefowl. *Behaviour* 127:309–322.
- Molenaar, R., S. de Vries, I. van den Anker, R. Meijerhof, B. Kemp, and H. van den Brand. 2010a. Effect of eggshell temperature and a hole in the air cell on the perinatal development and physiology of layer hatchlings. *Poult. Sci.* 89:1716–1723.
- Molenaar, R., R. Meijerhof, I. van den Anker, M. J. W. Heetkamp, J. J. G. C. van den Borne, B. Kemp, and H. van den Brand. 2010b. Effect of eggshell temperature and oxygen concentration on survival rate and nutrient utilization in chicken embryos. *Poult. Sci.* 89:2010–2021.
- Moran, E. T. Jr. 2007. Nutrition of the developing embryo and hatchling. *Poult. Sci.* 86:1043–1049.
- Müller, W., C. Eising, C. Dijkstra, and T. G. G. Groothuis. 2004. Within-clutch patterns of yolk testosterone vary with the onset of incubation in black-headed gulls. *Behav. Ecol.* 15:893–897.
- Piestun, Y., Y. Shinder, M. Ruzal, O. Halevy, and S. Yahav. 2008. The effect of thermal manipulations during the development of the thyroid and adrenal axes on in-hatch and post hatch thermoregulation. *J. Therm. Biol.* 33:413–418.
- Reis, L. H., L. T. Gama, and M. Chaveiro Soares. 1997. Effects of short storage conditions and broiler breeder age on hatchability, hatching time, and chick weights. *Poult. Sci.* 76:1459–1466.
- Ricklefs, R. E. 1987. Comparative analysis of avian embryonic growth. *J. Exp. Zool. Suppl.* 1:309–323.
- SAS Institute. 2004. SAS/STAT User's Guide. Version 9.1. SAS Inst. Inc., Cary, NC.
- Shinder, D., M. Rusal, M. Giloh, and S. Yahav. 2009. Effect of repetitive acute cold exposures during the last phase of broiler embryogenesis on cold resistance through the life span. *Poult. Sci.* 88:636–646.
- Tazawa, H., A. H. J. Visschedijk, J. Wittmann, and J. Piiper. 1983. Gas exchange, blood gases and acid-base status in the chick before, during and after hatching. *Respir. Physiol.* 53:173–185.
- Tona, K., R. D. Malheiros, F. Bamelis, C. Careghi, V. M. B. Moraes, O. Onagbesan, E. Decuyper, and V. Bruggeman. 2003. Effects of storage time on incubating egg gas pressure, thyroid hormones, and corticosterone levels in embryos and on their hatching parameters. *Poult. Sci.* 82:840–845.
- Tona, K., O. Onagbesan, V. Bruggeman, L. De Smit, D. Figueiredo, and E. Decuyper. 2007. Non-ventilation during early incubation in combination with dexamethasone administration during late incubation: 1. Effects on physiological hormone levels, incubation duration and hatching events. *Domest. Anim. Endocrinol.* 33:32–46.
- van de Ven, L. J. F., A. V. van Wagenberg, P. W. G. Groot Koerkamp, B. Kemp, and H. van den Brand. 2009. Effects of a combined hatching and brooding system on hatchability, chick weight, and mortality in broilers. *Poult. Sci.* 88:2273–2279.
- Wineland, M. J., V. L. Christensen, I. Yildrum, B. D. Fairchild, D. T. Ort, and K. M. Mann. 2006. Incubator environment interacts with genetic line of broiler at the plateau stage to affect embryo plasma thyroxine and triiodothyronine concentrations. *Int. J. Poult. Sci.* 5:714–722.
- Yalçın, S., V. Bruggeman, J. Buyse, E. Decuyper, M. Çabuk, and P. B. Siegel. 2009. Acclimation to heat during incubation: 4. Blood hormones and metabolites in broilers exposed to daily high temperatures. *Poult. Sci.* 88:2006–2013.