PRODUCTION, MODELING, AND EDUCATION

The effects of different eggshell temperatures on embryonic development, hatchability, chick quality, and first-week broiler performance

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ABSTRACT The aim of the current study was to determine the effects of different eggshell temperatures (EST) during 10 to 18 d of incubation on embryonic development, hatchability, chick quality, first-week broiler performance. The EST were maintained within the temperature ranges of 33.3 to 36.7, 37.8 to 38.2, and 38.9 to 40.0°C for the low, control, and high EST treatments, respectively. From d 15 to 18, embryo weight and relative embryo weight were found to be similar in the low and high EST groups. Salable chicks and hatchability of total eggs was found to be higher in the control EST group. Between d 10 and 17 of incubation, embryonic mortality in low, control, and high EST groups was determined to be 1.6, 0.8, and 2.0%, respectively. From d 18 to hatch, embryonic mortality and rate of dead and cull chicks were found to be significantly different. Hatching in high EST group was completed 26 h early, although hatching in low EST group was completed 10 h later than the control EST group. On the of hatching day, chick weight and length were found to be 39.5, 41.0, and 42.5 g, and 18.5, 21.4, and 19.1 cm in low, control, and high EST groups, respectively. The highest residual yolk sac weight and relative residual yolk sac weight were observed in high EST group as 7.7 g and 18.7%. Yolk-free chick weight and relative yolk-free chick weight were highest in the control EST group. At 1 wk of age, the BW in low, control, and high EST groups were determined as 131.1, 140.0, and 140.8 g, respectively. No significant difference was found for feed intake and feed conversion among treatments for wk 1. The mortality during the first week did not differ among groups; however, a higher mortality rate was observed numerically in the high EST group. In conclusion, embryo development, incubation parameters, chick quality, and the first week performance are affected by small changes in the EST.

Key words: incubation, eggshell temperature, embryonic development, broiler

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INTRODUCTION

An increase of 1 unit in the hatchability of total eggs, a primary criterion of productivity in breeder farms, converts into a great financial value over time (Ipek et al., 2004). To obtain optimum incubation results, the conditions during incubation must be adjusted to meet the requirements of the embryo (Meijerhof, 2009a). It has been well established that the embryonic environment influences the growth of the embryo in many species (Meijerhof, 2000; Hammond et al., 2007; Leksrisompong et al., 2007).

The process of converting the content of an egg into a 1-d-old chick is driven by temperature (Meijerhof, 2009b). Optimum incubation temperature is normally defined as that required to achieve maximum hatchability (Wilson, 1991; Hulet et al., 2007; Shim and Pesti, 2011). French (2000) reported that even a small temperature difference can have a significant effect on embryonic development. Deviations from optimum incubation temperatures may affect embryo size, organ and skeletal growth, and hatching success (Yalcın and Siegel, 2003; Tazawa et al., 2004). The temperature within the egg [i.e., the embryo temperature (Meijerhof, 2009a)] is particularly critical, and maintaining the correct embryonic temperature during incubation has been shown to be more important than the incubator temperature settings (Meijerhof, 2009a). Therefore, trying to control embryo temperatures between acceptable ranges will result in a better hatchability and better chick quality (Meijerhof, 2009a). If the incubation temperature is too low or too high (34.6°C vs. 40.6°C), embryonic mortality will be increased, and therefore hatchability will be decreased (Decuypere et al., 1979; Suarez et al., 1996; Willemsen et al., 2010). In practice, deviations of over 4°C have been found, depending on location in the incubator and embryonic age (Lourens, 2001; Joseph et al., 2006).

The effects of temperature on length of incubation (Michels et al., 1974; French, 1994; Shim and Pesti, 2011) and on the rate of embryo growth (Decuypere et

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al., 1979) have been observed in several studies. Wineland et al. (2000) demonstrated that differences in embryo temperature in the setter and hatcher resulted in a difference in the development of the whole chicken and specific organs. The heart has been the organ most consistently affected by abnormal incubation temperature (Christensen et al., 2004; Leksrisompong et al., 2007; Shim and Pesti, 2011). Differences in embryo temperature can vary development of embryo and the quality of the hatched chick (Lourens, 2001; Hammond et al., 2007; Meijerhof, 2009b).

It has been demonstrated that a low eggshell temperature (**EST**) from 0 to 10 d of incubation (36.6°C), which may arise in practice, reduced embryonic weight, hatchability, and early chick quality (Joseph et al., 2006). In contrast, chick embryos respond to elevated incubation temperature (38.7 to 39.7°C) from d 16 of incubation to hatching day with accelerated growth and development (Hulet et al., 2007). Leksrisompong et al. (2007) found similar results when they applied higher incubation temperatures (39.5°C) after d 14 of incubation. However, this accelerated development negatively affected hatchling chick weight and increased the number of cull chicks (Joseph et al., 2006; Molenaar et al., 2011).

The developing embryo uses the content of the egg to develop its body (Meijerhof, 2009a). For this process, energy is essential and is mainly derived from using the fat sources in the yolk. Embryos use the nutrients from the yolk sac to initiate body growth (Chamblee et al., 1992; Murakami et al., 1992; Meijerhof, 2009a), development of the small intestine (Noy and Sklan, 1999), and other organs. Moreover, withdrawal of the yolk sac into the abdomen of the embryo before hatching provides nutrients to the newly hatched avian neonate during the first few days of life. The residual yolk sac comprises approximately 14% of the chick BW at the time of hatching (Mikec et al., 2006; Meijerhof, 2009a). The 1-d-old chick weight includes the actual chick weight and the weight of the residual yolk sac. If a large amount of residual yolk sac remains at hatch, this means that less of this energy source has been used during incubation and the chick less developed (Preez, 2007).

The quality of the 1-d-old chick is important for a good start by the chick will be and for the final performance of the bird (Meijerhof, 2009b). Wolanski et al. (2004) showed that the length of the chicken is indicative for its development and a positive correlation between chick length at day of hatch and broiler performance existed (Meijerhof, 2009b). Several studies have shown that embryo temperature deviations also influenced posthatch broiler performance and processing yields (Wilson, 1991; Lourens and van Middelkoop, 2000). Gladys et al. (2000) showed that a 2°F difference in embryo temperature resulted in a significant difference in embryo growth and the feed conversion of broilers at 6 wk of age.

Studies on this subject have been usually performed to compare the effects of lower or higher egg shell temperatures to control temperatures during early incubation (4–7 d; Joseph et al., 2006; Shim and Pesti, 2011) or late incubation (18–21 d; Hulet et al., 2007; Leksrisompong et al., 2007; Willemsen et al., 2010) on different parameters (e.g., bone development and leg problems, embryo development, organ development, incubation parameters, chick quality). The aim of the current study was determine the effects of different egg shell temperatures (low, control, and high) during 10 to 18 d of incubation on embryonic development, hatchability, chick quality, and first-week broiler performance. In the study, embryo development and yolk sac absorption were monitored daily during incubation and also for 3 d after hatching.

MATERIALS AND METHODS

Experimental Design

A total of 1,800 eggs were obtained from a commercial Cobb 500 broiler breeder parent stock at 35 wk of age, were stored at 16°C and 65% RH for 2 d, and warmed to room temperature (22°C) for 8 h before setting. All eggs were numbered, weighed (55–60 g) before incubation, and then incubated in the same incubator (1,800 capacity egg setter, T2400 C, Cimuka Inc., Ankara, Turkey) at 37.5°C and a RH of 55 to 60% during the first 10 d of incubation. On d 10 of incubation, the eggs were separated into 3 groups and incubated in fully automated ventilation, programmable incubators at full capacity (600 capacity egg setter, 6 trays; T640 I, Cimuka Inc.).

The eggshell temperature was measured by contact at the equator of the egg using an infrared digital thermometer (IRT 4520, Thermoscan, Braun, Germany) with a total of 60 eggs per treatment (10 eggs per each tray) from embryonic d 10 to 18. The incubator temperatures were ignored, and the eggshell temperature was considered. The incubator temperatures were programmed daily based on the eggshell temperature. The EST was maintained for the 3 groups: low EST, control EST, and high EST within the ranges of 33.3 to 36.7, 37.8 to 38.2, and 38.9 to 40.0°C, respectively. In this study, the negative effects of excessively high temperatures on incubation results were considered important. Therefore, high EST treatment was applied in a narrower temperature range. The infrared thermometer was allowed to equilibrate on the floor of an incubator for 10 min before each use. The EST was measured after opening the incubator and then the incubator door was closed.

From a total of 15 eggs per treatment group, embryos were killed daily by cervical dislocation, weighed, and samples taken from embryonic d 12 to hatching day (Willemsen et al., 2010). Embryos from each treatment group were measured for embryo weight and yolk sac

weight (Torres et al., 2012). Eggs used for sampling were weighed before opening to calculate egg weight loss (Fasenko et al., 2009). After embryos were excised from the extra embryonic membranes, they were carefully separated from the yolk sac and excessive fluid was dried off with absorbent paper. Embryos and yolk sacs were weighed to calculate relative embryo and yolk weights.

$$\text{Relative embryo weight (\%)} = (\text{yolk-free embryo weight/egg weight at setting}) \times 100.$$

$$\text{Relative yolk weight (\%)} = (\text{yolk weight/egg weight at setting}) \times 100.$$

On d 18y of incubation, eggs were transferred to incubator (T2400 C, Cimuka Inc., Ankara). After transfer to a hatching, the number of hatched chicks started to be counted from 456 to 518 h of incubation at regular 6-h intervals and the incubation period determined for treatment groups (Collin et al., 2005).

At hatch, chicks were classified as salable (clean, dry, and without deformities) or culls (splayed legs, unhealed navels, and so on; Tona et al., 2004; Molenaar et al., 2011). The percentage of salable and cull chicks was expressed as a percentage of fertile eggs (Molenaar et al., 2011). Unhatched eggs were opened to macroscopically determine fertility and embryonic mortality (early, middle, late). Saleable chicks and hatchability of total eggs were calculated. Fertility was calculated as the ratio of total eggs at set to fertile eggs. Chick hatch weight was determined by weighing all chicks hatched individually.

After hatching, all chicks were weighed at feather dryness (approximately 2 h posthatch). A group of 30 chicks from each group was randomly sampled to determine the cloacal temperature. The cloacal temperatures of the chicks were also measured (to the nearest 0.01°C) using a thermocouple thermometer that was inserted into the cloaca. These chicks from each temperature treatment were killed by cervical dislocation to determine chick weight and length, residual yolk sac weight, and yolk-free chick weight. Chick length was measured only for the first day from the tip of the beak to the tip of the longest toe by placing the chick face down on a flat surface and straightening the left leg. The other measurements were repeated for 3 d posthatch.

$$\text{Relative chick weight (\%)} = (\text{yolk-free chick weight/chick weight}) \times 100.$$

$$\text{Relative residual yolk sac weight (\%)} = (\text{residual yolk sac weight/chick weight}) \times 100.$$

The chicks (n = 720) were randomly allocated into treatment groups (low, control, and high EST), after completing hatching in each of the groups. The chicks were placed in 18 floor pens with a floor space of 2.0 × 2.0 m^2 to provide 6 replicate pens and 40 chicks per pen. The chicks received a standard crumbled broiler starter diet (22.5% CP and ME 3,057 kcal/kg) between d 1 to 7 and were exposed to 24 h of light for the first week. Feeding was supplied by plastic hanging feeders. Water was supplied to both groups with round type drinkers for ad libitum consumption and was regularly refreshed. The temperature, humidity, and other environmental factors were equal for each of the groups during the trial. The live BW gain values were monitored at the end of the first week and feed conversion were calculated using the feed intake and BW gain values. The mortality was recorded daily during the first week.

In this study, the care and use of animals were in accordance with the laws and regulations of Turkey and approved by the Ethical Committee of the Uludağ University (License number 2012–01/02).

Statistical Analyses

Data were subjected to ANOVA (1989, SAS Institute Inc., Cary, NC), utilizing ANOVA procedures for balanced data. Analysis for percentage data were conducted after square root of arc sine transformation of the data. Significant differences among treatment means were determined by Duncan's multiple range test. Mortality data were analyzed using chi-squared tests.

RESULTS

The effects of different EST treatments on yolk sac weight and relative yolk sac weight are presented in Table 1. From d 12 of incubation to d 15, there was no significant difference among treatment groups. However, on d 15 and 16 of incubation, the highest yolk sac weight and relative yolk sac weight were found in the high EST group, from d 17 to 21, the lowest yolk sac weight and relative yolk sac weight was found in the high EST group because of increased yolk absorption during those days ($P < 0.05$).

The effects of EST treatments on embryo weight and relative embryo weight are presented in Table 2. Between d 12 and 14, the lowest embryo weight and relative embryo weight were found in the low EST group ($P < 0.05$). However, from d 15 to 18, embryo weight and relative embryo weight were found to be similar in low and high EST groups; these parameters were highest in the control EST group from d 12 to 18 ($P < 0.05$). After d 18, the high EST group embryo weight increased faster, and in this group hatching was completed on d 21 of incubation.

The effects of treatments on incubation results are presented in Table 3. The effects of different EST on egg weight and fertility were not significant as was expected. Salable chicks and hatchability of total eggs were higher in the control EST group than others ($P <$

Table 1. Mean values of the yolk sac weight (g) and relative yolk sac weight (%) for eggshell temperature (EST) groups[1]

Day of incubation	Yolk sac weight (g)				Relative yolk sac weight (%)			
	Low EST	Control EST	High EST	*P*-value	Low EST	Control EST	High EST	*P*-value
12	17.8 ± 0.40	17.3 ± 0.82	17.9 ± 0.53	0.863	30.3 ± 0.51	29.3 ± 0.33	30.4 ± 0.45	0.884
13	16.8 ± 0.38	16.7 ± 0.43	16.7 ± 0.27	0.782	28.5 ± 0.42	28.3 ± 0.54	28.2 ± 0.31	0.865
14	13.5 ± 0.46	13.1 ± 0.21	13.4 ± 0.23	0.784	22.9 ± 0.78	22.1 ± 0.40	22.6 ± 0.52	0.873
15	12.7 ± 0.41^{b}	12.2 ± 0.32^{c}	13.0 ± 0.25^{a}	0.041	21.6 ± 0.41^{b}	20.7 ± 0.51^{c}	22.1 ± 0.51^{a}	0.043
16	12.1 ± 0.30^{b}	11.7 ± 0.56^{c}	12.9 ± 0.23^{a}	0.027	20.5 ± 0.35^{b}	19.9 ± 0.45^{c}	21.9 ± 0.38^{a}	0.037
17	11.7 ± 0.39^{a}	11.1 ± 0.26^{b}	10.5 ± 0.28^{c}	0.034	19.9 ± 0.45^{a}	18.8 ± 0.24^{b}	17.8 ± 0.31^{c}	0.041
18	11.7 ± 0.69^{a}	10.6 ± 0.45^{b}	10.1 ± 0.32^{c}	0.021	19.9 ± 0.75^{a}	18.0 ± 0.53^{b}	17.0 ± 0.41^{c}	0.037
19	9.7 ± 0.46^{A}	8.4 ± 0.28^{B}	7.3 ± 0.43^{C}	0.001	16.5 ± 0.58^{A}	14.3 ± 0.35^{B}	12.4 ± 0.48^{C}	0.001
20	6.5 ± 0.33^{A}	5.4 ± 0.45^{B}	2.3 ± 0.28^{C}	0.001	11.0 ± 0.51^{A}	9.2 ± 0.51^{B}	3.9 ± 0.51^{C}	0.001
21	2.3 ± 0.52^{A}	2.2 ± 0.75^{A}	0.00 ± 0.00^{B}	0.001	5.78 ± 1.34^{A}	5.25 ± 1.72^{A}	0.00 ± 0.00^{B}	0.001

$^{a–c}$Means ± SEM in a row with different superscripts differ significantly ($P < 0.05$).

$^{A–C}$Means ± SEM in a row with different superscripts differ significantly ($P < 0.01$).

[1]For yolk sac weight (g) and relative yolk sac weight (%), a total of 15 embryos from each group for each day were randomly sampled.

Table 2. Mean values of the embryo weight (g) and relative embryo weight (%) for eggshell temperature (EST) groups[1]

Day of incubation	Embryo weight (g)				Relative embryo weight (%)			
	Low EST	Control EST	High EST	*P*-value	Low EST	Control EST	High EST	*P*-value
12	6.3 ± 0.24^{c}	7.2 ± 0.39^{a}	6.9 ± 0.27^{b}	0.038	10.7 ± 0.30^{c}	12.3 ± 0.41^{a}	11.7 ± 0.35^{b}	0.040
13	6.9 ± 0.21^{c}	7.6 ± 0.38^{a}	7.2 ± 0.29^{b}	0.032	11.7 ± 0.33^{c}	12.9 ± 0.45^{a}	12.2 ± 0.41^{b}	0.036
14	7.3 ± 0.37^{c}	8.5 ± 0.49^{a}	7.8 ± 0.35^{b}	0.024	12.4 ± 0.54^{c}	14.4 ± 0.57^{a}	13.3 ± 0.49^{b}	0.033
15	9.8 ± 0.50^{b}	11.8 ± 0.54^{a}	10.0 ± 0.48^{b}	0.037	16.7 ± 0.54^{b}	20.0 ± 0.61^{a}	17.0 ± 0.51^{b}	0.041
16	13.4 ± 0.83^{b}	15.7 ± 0.62^{a}	14.0 ± 0.80^{b}	0.028	22.8 ± 0.90^{b}	26.6 ± 0.73^{a}	23.6 ± 0.85^{b}	0.032
17	16.1 ± 0.63^{b}	18.4 ± 0.61^{a}	16.5 ± 0.59^{b}	0.037	27.3 ± 0.99^{b}	31.2 ± 0.99^{a}	27.9 ± 0.99^{b}	0.040
18	19.1 ± 0.61^{b}	21.0 ± 0.71^{a}	19.5 ± 0.54^{b}	0.024	32.4 ± 0.77^{b}	35.6 ± 0.73^{a}	33.0 ± 0.69^{b}	0.030
19	22.9 ± 0.52^{C}	25.2 ± 0.95^{B}	30.3 ± 0.75^{A}	0.001	39.0 ± 0.59^{C}	42.8 ± 0.98^{B}	51.3 ± 0.81^{A}	0.001
20	26.3 ± 0.62^{C}	30.4 ± 0.74^{B}	37.3 ± 0.75^{A}	0.001	44.8 ± 0.74^{C}	51.6 ± 0.77^{B}	63.2 ± 0.81^{A}	0.001
21	37.1 ± 0.64^{C}	40.1 ± 0.42^{B}	40.8 ± 0.08^{A}	0.001	64.6 ± 1.86^{C}	68.8 ± 1.99^{B}	71.9 ± 2.53^{A}	0.001

$^{a–c}$Means ± SEM in a row with different superscripts differ significantly ($P < 0.05$).

$^{A–C}$Means ± SEM in a row with different superscripts differ significantly ($P < 0.01$).

[1]For embryo weight (g) and relative embryo weight (%), a total of 15 embryos from each group for each day were randomly sampled. Relative embryo weight (%) = (yolk-free embryo weight/egg setting weight) × 100.

0.01). Between d 1 and 10, there were no differences for embryonic mortality. Between d 10 and 17, embryonic mortality in the low, control, and high EST groups were determined as 1.6, 0.8, and 2.0%, respectively, and the differences among the groups were significantly different ($P < 0.01$). From d 18 to hatching day, embryonic mortality and rate of dead, cull chicks were found to be significant among low, control, and control EST groups as 7.2, 1.6, and 7.8% and 2.7, 0.9, and 2.7%, respectively ($P < 0.01$). However, egg weight loss was lowest in the low EST group (11.00%) and highest in the high EST group (13.8%; $P < 0.05$). The effects of different EST treatments on chick hatch weight was found to be significant ($P < 0.05$). Chick hatch weight was determined as 39.2, 42.3, and 41.1 g in the low, control, and high EST groups, respectively. Chick weight/initial egg weight ratio was found to be significantly different among groups and highest in the control EST group and lowest in the low EST group ($P < 0.05$). Incubation length varied at 518, 508, and 482 h in the low, control, and high EST groups, respectively. Hatching in the high EST group was completed 26 h earlier than the low EST group, and hatching in the low EST group was completed 10 h later than control EST group. Although, the cloacal temperature was found to be higher numerically (40.5°C) in the high EST group, as was expected, it was not significantly different among the groups ($P = 0.058$).

The effects of different EST on chick weight, chick length, residual yolk sac weight, relative residual yolk sac weight, yolk-free chick weight, and relative yolk-free chick weight are presented in Table 4. On the first day, chick weight and length were found as 39.5, 42.5, and 41.0 g and 18.5, 21.4, and 19.1 cm in the low, control, and high EST groups, respectively ($P < 0.05$). The highest residual yolk sac weight and relative residual yolk sac weight were observed in the high EST group as 7.7 g and 18.7% ($P < 0.01$). Although yolk-free chick weight was found to be the highest in the control EST group as 36.2 g, relative yolk-free chick weight was found to be similar in the low (87.2%) and control (85.3%) EST groups ($P < 0.05$). On the second day, there was a significant difference among treatment groups for chick weight, the lowest chick weight was found in the low EST group as 47.2 g ($P < 0.05$). On d 2, residual yolk sac weight and relative residual yolk sac weight were greatest in the high EST group and lowest residual yolk sac weight and relative residual yolk sac weight were found in control EST group ($P < 0.05$). However, relative yolk-free chick weight was found to be highest in the control EST group and lowest in the high EST group. In the low, control, and high EST groups, relative yolk-free chick weight was 91.0, 94.4, and 89.4, respectively ($P < 0.05$). On the third day, the lowest chick weight was also found in the low EST group at 52.2 g ($P < 0.05$). Residual yolk sac weight and relative residual yolk sac weight were the highest in the high EST group (4.8 g, 8.6%) and lowest in the control EST group (1.6 g, 2.8%; $P < 0.05$). Yolk-free chick weight and relative yolk-free chick weight were the highest in the control EST group (54.7 g; 97.2%), and there were no significant differences for these parameters between the low and high EST groups ($P < 0.05$).

The effects of different EST on the posthatch first-week broiler performance parameters are presented in Table 5. The initial BW on d 1 were similar for the low EST (39.6 g) and high EST groups (41.0 g). The control EST group (42.4 g) was heavier ($P = 0.024$). There was a significant difference in the live BW during the first week. At 1 wk of age, the BW and growth rate in the low, control, and high EST group were determined as 131.1, 140.0, 140.8 g, and 91.5, 97.6, and 99.8 g, respectively, and the low EST group was found to be lighter than others ($P < 0.05$). No significant difference for feed intake and feed conversion among treatments for the first week was observed. The mortality during

Table 3. The effects of eggshell temperature (EST) on incubation results and cloacal temperature[1]

	EST			
Characteristic	Low EST	Control EST	High EST	*P*-value
Egg weight (g)	58.8 ± 1.20	58.93 ± 1.10	59.0 ± 0.90	0.767
Fertility (%)	96.9 ± 0.63	97.2 ± 0.87	96.5 ± 1.31	0.832
Salable chicks (%)	86.9 ± 1.72^{B}	95.4 ± 1.98^{A}	86.4 ± 1.14^{B}	0.001
Hatchability of total eggs (%)	86.9 ± 1.26^{B}	94.0 ± 2.14^{A}	86.0 ± 1.90^{B}	0.001
1–10 d embryos died (%)	1.6 ± 0.50	1.3 ± 1.50	1.1 ± 1.06	0.664
10–17 d embryos died (%)	1.6 ± 0.40^{B}	0.8 ± 0.35^{C}	2.0 ± 0.48^{A}	0.001
18–21 d embryos died (%)	7.2 ± 1.24^{A}	1.6 ± 0.85^{B}	7.8 ± 1.82^{A}	0.001
Dead + cull chicks (%)	2.7 ± 0.28^{A}	0.9 ± 0.26^{B}	2.7 ± 0.17^{A}	0.001
Egg weight loss (%)	11.0 ± 1.07^{c}	12.3 ± 1.18^{b}	13.8 ± 1.12^{a}	0.042
Chick hatch weight (g)	39.2 ± 1.20^{b}	42.3 ± 1.00^{a}	41.1 ± 1.00^{b}	0.015
Chick weight/initial egg weight (%)	66.7 ± 1.40^{c}	71.8 ± 1.45^{a}	69.6 ± 1.34^{b}	0.035
Incubation length (h)	518	508	482	
Cloacal temperature (°C)	38.7 ± 1.70	39.4 ± 1.50	40.5 ± 1.00	0.058

$^{a–c}$Means ± SEM in a row with different superscripts differ significantly ($P < 0.05$).

$^{A–C}$Means ± SEM in a row with different superscripts differ significantly ($P < 0.01$).

[1]For chick hatch weight, all chicks were weighed individually, and for cloacal temperature, a total of 30 chicks from each group were randomly sampled. Egg weight loss (%) = (egg setting weight − egg transfer weight) × 100/egg setting weight.

Table 4. Mean values of the chick weight (g), chick length (cm), residual yolk sac weight (g), relative residual yolk sac weight (%), yolk-free chick weight (g), and relative yolk-free chick weight (%) for eggshell temperature (EST) groups[1]

Item	Chick weight (g)	Chick length (cm)	Residual yolk sac weight (g)	Relative residual yolk sac weight (%)	Yolk-free chick weight (g)	Relative yolk-free chick weight (%)
1 d posthatch						
Low EST	39.5 ± 1.50^{b}	18.5 ± 1.40^{b}	5.1 ± 0.61^{C}	12.9 ± 1.11^{C}	34.5 ± 1.30^{b}	87.2 ± 1.51^{a}
Control EST	42.5 ± 1.10^{a}	21.4 ± 1.60^{a}	6.3 ± 0.92^{B}	14.7 ± 1.30^{B}	36.2 ± 1.50^{a}	85.3 ± 1.60^{a}
High EST	41.0 ± 1.10^{b}	19.1 ± 1.41^{b}	7.7 ± 1.20^{A}	18.7 ± 1.30^{A}	33.4 ± 1.21^{b}	81.3 ± 1.32^{b}
P-value	0.021	0.044	0.001	0.001	0.036	0.018
2 d posthatch						
Low EST	47.2 ± 1.72^{b}	—	4.2 ± 0.52^{b}	9.0 ± 0.84^{b}	43.0 ± 2.18^{b}	91.0 ± 2.84^{b}
Control EST	50.2 ± 2.12^{a}	—	2.8 ± 0.71^{c}	5.6 ± 0.80^{c}	47.4 ± 2.46^{a}	94.4 ± 3.08^{a}
High EST	51.0 ± 4.21^{a}	—	5.4 ± 0.44^{a}	10.6 ± 0.57^{a}	45.6 ± 3.49^{ab}	89.4 ± 3.54^{c}
P-value	0.035	—	0.018	0.012	0.042	0.045
3 d posthatch						
Low EST	52.2 ± 0.77^{b}	—	3.2 ± 0.82^{b}	6.1 ± 0.90^{b}	49.0 ± 1.33^{b}	93.9 ± 2.04^{b}
Control EST	56.3 ± 1.75^{a}	—	1.6 ± 0.55^{c}	2.8 ± 0.63^{c}	54.7 ± 1.91^{a}	97.2 ± 2.47^{a}
High EST	55.3 ± 2.71^{a}	—	4.8 ± 0.45^{a}	8.6 ± 0.57^{a}	50.5 ± 2.05^{b}	91.4 ± 2.80^{b}
P-value	0.041		0.012	0.019	0.033	0.042

[a–c]Means ± SEM in a row with different superscripts differ significantly ($P < 0.05$).

[A–C]Means ± SEM in a row with different superscripts differ significantly ($P < 0.01$).

[1]For chick weight, chick length, residual yolk sac weight, relative residual yolk sac weight, yolk-free chick weight, and relative yolk-free chick weight, a total of 15 chicks from each group for each day were randomly sampled.

first week did not differ among the EST treatments (chi-square = 4.576, $P = 0.101$).

DISCUSSION

Studies have shown that incubation conditions influence embryo development (Freeman and Vince, 1974; Decuypere and Michels, 1992; Lourens et al., 2005, 2007; Molenaar et al., 2011), and incubation temperature is one of the most important physical factors influencing embryo development (Decuypere and Michels, 1992; Meijerhof, 2000; Lourens, 2001; Leksrisompong et al., 2007; Willemsen et al., 2010). Studies have shown that lower incubation temperatures (35°C) after embryonic d 14 slowed embryonic growth and increased the incubation period (Black and Burggren, 2004). In contrast, higher incubation temperatures (39.5°C) after embryonic d 14 accelerated embryonic growth and development (Leksrisompong et al., 2007).

In this study, where embryo weight and relative embryo weight were found to be highest in the control EST group from d 15 to 18, after d 19 embryo weight increased faster and hatching was completed on d 20 in the high EST group. The results of this study agree with those of previous studies (Ricklefs, 1987; Suarez et al., 1996; Black and Burggren, 2004). In another similar study, Shim and Pesti (2011) found that incubation periods were +17 h for 36.5°C eggs and −10 h for 38.5°C compared with the incubation period at 37.5°C (508 h).

The yolk sac is of a vital importance for embryo development and has a highly vascularized membrane that starts to develop and surround the yolk at around d 2 of incubation (Meijerhof, 2009a). Also, the absorption of nutrients from the yolk sac is essential to initiate body growth (Chamblee et al., 1992; Murakami et al., 1992; Meijerhof, 2009a) and for development of the small intestine (Noy and Sklan, 1999). If there was a large amount of residual yolk, less development had occurred and the embryo growth should not be considered as optimal (Meijerhof, 2009b). In this study, hatching in the high EST group was completed by d 20 of incubation and it was found that the highest yolk sac absorption and also embryo weight were observed in the high EST group on this day. It clearly showed

Table 5. Mean values of the posthatch first-week broiler performance parameters for eggshell temperature (EST) groups[1]

Broiler performance parameter	Low EST	Control EST	High EST	*P*-value
Initial weight (g/bird)	39.6 ± 1.81^{b}	42.4 ± 1.23^{a}	41.0 ± 1.30^{b}	0.024
BW (g/bird)	131.1 ± 3.33^{b}	140.0 ± 3.40^{a}	140.8 ± 2.51^{a}	0.032
Growth rate (g/bird)	91.5 ± 2.82^{b}	97.6 ± 3.21^{a}	99.8 ± 4.10^{a}	0.037
Feed intake (g/bird)	108.8 ± 3.41	106.5 ± 3.22	104.9 ± 2.31	0.602
Feed conversion	1.19 ± 0.18	1.09 ± 0.15	1.05 ± 0.12	0.842
Mortality (chi-square = 4.576)	1.66 (4/240)	0.42 (1/240)	2.91 (7/240)	0.101

[a,b]Means ± SEM in row with different superscripts differ significantly ($P < 0.05$).

[1]Feed conversion = feed intake/BW gain.

that absorption of higher amount of yolk sac increased embryo weight. In the high EST group, embryo weight increased strikingly more than other groups. This can be explained by embryo water intake accelerating before hatching. Therefore, when the high EST group was compared with low and control EST groups for embryo weights on the prehatching day, higher embryo weight was observed in the control EST group than in others.

Practical experience and scientific research shows that controlling embryo temperatures between acceptable ranges results in better hatchability and better chick quality (Meijerhof, 2009b). If the incubation temperature is too low or too high during different incubation periods, embryonic mortality will be increased, and therefore hatchability and chick quality will be decreased (Decuypere et al., 1979; Suarez et al., 1996; Lourens, 2001; Joseph et al., 2006; Willemsen et al., 2010). Other studies have shown that a high EST in the second half of incubation can increase embryonic mortality in the last week of incubation (French, 1994; Lourens et al., 2005; Willemsen et al., 2010; Molenaar et al., 2011). Contrary to the findings, several researchers concluded that embryonic mortalities were not influenced by incubation temperature (Yalcın et al., 2010; Shim and Pesti, 2011). Molenaar et al. (2011) found that eggshell temperature did not affect hatchability, but a high EST increased the percentage of second-grade chickens by 0.7%. In this study, results show that lower and higher eggshell temperatures can increase the number of cull chicks. Although RH was similar (55–60%) among the experimental groups, the effects of EST on egg weight loss were found to be significant. In the high EST group, a higher egg weight loss was observed because of increasing water loss. Body weight variability at hatch (when egg weight was constant) was often the result of differences in moisture loss or residual yolk sac weight (Tullett and Burton, 1982; Joseph et al., 2006). In another study, BW was lower in chicks incubated at higher temperature compared with those incubated at lower temperature (44.7 vs. 42.9 g). Contrary to our result, it was found that BW was lower (49.0 g) at 36.5°C than at 37.5°C (49.6 g) or 38.5°C (50.6 g; Shim and Pesti, 2011).

The quality of the 1-d-old chick has been demonstrated to be important for a good start for the chick and for broiler performance (Meijerhof, 2009b). Before hatching, absorption of the yolk sac into the abdomen of the embryo provides nutrients for the chicks during the first few days of life. Chick weight has been measured as a chick quality criterion and is a combination of the real chick weight and the remaining yolk residual. In this study, on the first day after hatching in the high EST group, chick weight composed approximately 18.7% of the residual yolk sac. On d 1, after subtracting yolk sac weight from chick weight, low EST chicks weighed the same as high EST chicks. On the second and third days, the low and high EST groups residual yolk sac was found to be greater in weight than the control EST group. However, yolk-free chick weight was found to be lower in weight. Therefore, chick weight differences between low and high EST treatments may be explained by differences in yolk sac weight (Joseph et al., 2006). These results agreed with other studies (Mikec et al., 2006; Preez, 2007). In other studies, high EST (≥38.9°C), as compared with normal EST (37.8°C), during the second half of incubation reduced hatchling quality as expressed by a lower yolk-free chick weight (Lourens et al., 2005; Hulet et al., 2007; Leksrisompong et al., 2007; Lourens et al., 2007; Molenaar et al., 2011). In contrast, Shim and Pesti (2011) found that BW was lower in chicks incubated at the higher temperature (38.5°C) compared with those incubated at the lower temperature (36.5°C) (44.7 vs. 42.9 g). Molenaar et al. (2011) found that BW at hatch was 3.4 g less and yolk-free chick weight was 3.0 g less in the high EST (38.9°C) treatment when compared with the normal EST (37.8°C) treatment.

Length of the chick was indicative for its development and a criterion of 1-d-old chick quality. It was stated that chick length has a substantially higher positive correlation with broiler performance than 1-d-old chick weight, especially when corrected for egg weight (Wolanski et al., 2004; Meijerhof, 2009b). Molenaar et al. (2007) showed that, when originating from eggs of equal weight, an increase in chick length at hatch resulted in an increased BW in male broilers. Several researchers also concluded that high EST (≥38.9°C) resulted in a shorter chick length (Hulet et al., 2007; Leksrisompong et al., 2007; Lourens et al., 2007; Molenaar et al., 2011). Conversely, there were findings that lower eggshell temperature (36.6°C) from 0 to 10 d of incubation reduced chick length (Joseph et al., 2006). In this study, chick length increased in parallel with increasing chick weight and was greatest at 21.4 cm in the control EST group.

As the initial BW of the low and high EST groups were similar, although the control group was heavier; after 1 wk posthatch, the effect of different eggshell temperatures on chick weight could be observed, where chicks from the low EST still weighed less than the others. Similarly, Shim and Pesti (2011) also reported that changes in the incubation temperature of as little as 1°C during embryonic d 4 to 7 affect chick initial BW and they found hatching chick weight of 49.0, 49.6, and 50.6 g in lower (36.5°C), control (37.5°C), and higher (38.5°C) incubation temperatures, respectively. However, Hammond et al. (2007) found that incubation at high temperatures increased the chick initial BW. In contrast to our findings, Yalcın et al. (2010) reported that chick weights were not influenced by incubation temperature.

In this study, different egg shell temperatures did not affect feed intake, feed conversion, and mortality at 7 d of age. The total mortality between 1 and 7 d of age

did not differ among the EST treatments. However, a higher numerical mortality rate was observed in the high EST group.

In conclusion, embryo development, incubation parameters, chick quality, residual yolk sac weight, and yolk-free chick weight were affected by small changes in the EST. Chick quality and first-week mortality have great importance for profitability of producers. In this study, higher number of cull chicks and shorter chick length, an indicator of chick quality, in low and high EST groups showed that eggshell temperature during incubation was very important.

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