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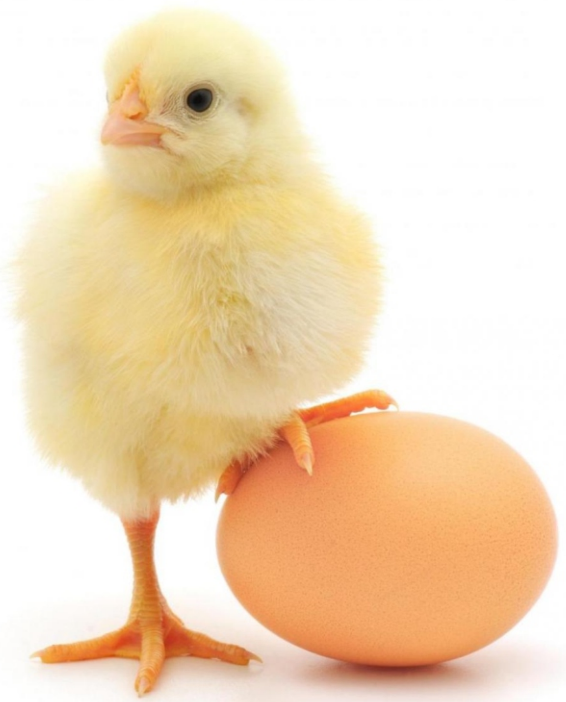

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Research Article
Relationship of Thermal Regulation Capacity of Chicks with Breeder age, Breed and Incubator type

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Abstract
Background and Objective: In chickens, development of the thermoregulatory system during incubation can increase thermal regulation efficiency after hatching and is critical to ensure animal welfare. Several factors, including breeder age, incubator type and breed can all affect thermoregulatory system development. This study evaluated how these factors are related to thermoregulatory system development and how they affect thermal regulation capacity of chicks. **Materials and Methods:** The experimental design was completely randomized, totaling 12 treatments with 6 replicates each. Each cart inside an incubator served as an experimental unit that contained 5,040 eggs and each type of incubator contained 36 carts. The eggs were derived from Cobb 500 and Ross breeds at three different ages. **Results:** Chicks produced by young Ross breeders and old Cobb breeders had a lower capacity to remain warm when incubated in a multiple stage incubator and while in the dispatch room. **Conclusion:** Breed type and breeder age can affect thermoregulatory system development of chicks. A comfortable ambient temperature is needed to ensure appropriate development of chicks.

Key words: Animal welfare, hatchery, incubation, poultry, thermoregulatory system

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Data Availability: All relevant data are within the paper and its supporting information files.

INTRODUCTION

Brazil is now the largest exporter and the second largest producer of chicken meat in the world. The poultry industry in Brazil represents 1.5% of GDP and generates 3.5 million direct and indirect jobs[1]. To maintain exports and reach new external markets, poultry companies need to adapt principles of animal welfare at all stages of poultry production, including in the hatchery.

There are few studies concerning animal welfare in the hatchery process despite the fact that inappropriate hatchery conditions can cause stress early in life that may have negative implications for poultry production, broiler performance and overall animal health. For example, thermal stress at the hatchery is associated with increased mortality of chicks up to 14 days old and low performance of broilers[2].

During the embryonic stage of chickens, the neural system develops and the thermoregulatory capacity is determined. Temperature regulation is critical between the embryonic stage and maturity, particularly soon after hatching[3]. The temperature in the hatchery is usually somewhat lower than the incubator temperature to reduce the risk of overheating soon after hatching. However, when the hatching time window is broad, or there is a long interval between hatching of the first and last chicks, the first chicks may overheat[4,5]. Chicks are also exposed to temperature challenges when they are separated from eggshell debris a period when they are subjected to intense handling that can cause stress and in turn hinder the body temperature control of the chick[4].

Soon after hatching and separation from egg shell debris, the chicks are vaccinated against Avian Infectious Bronchitis by spraying. During this process, decreases in chick body temperature can induce thermal shock or even hypothermia, particularly since chicks have high energy expenditure due to stress during processing. In addition, if the ambient temperature in the dispatch room is below the comfort temperature of 32°C or if the boxes are wet, cold sensations by the chicken intensify[4].

Therefore, understanding how the thermal regulation ability of a chick is influenced by external conditions is important to ensure poultry productivity. The objective of this study was to evaluate the relationship between thermal regulation capacity of chicks and breed, breeder age and incubator type.

MATERIALS AND METHODS

This project was approved by the Ethics Committee on the Use of Animals of the UFPR, Sector Palotina city (CEUA/Palotina) under protocol no. 19/2017. The experiment was carried out in November 2017 in a commercial hatchery in the western region of the state of Paraná. The eggs were from the farm of the same cooperative (company).

The incubation parameters used were:

- **Multiple stage incubator:** 98.7°F and 83% relative humidity (RH)
- **Multiple stage hatcher trays:** 98.5°F and 84% RH
- **Single stage incubator:** 97.4°F-100.5°F;40%-70% RH; 3,500-10,000 ppm CO_2
- **Single stage hatcher trays:** 96.0°F- 98.2°F, 40%-68% RH; 3,200-4,000 ppm CO_2.

The experimental design was completely randomized in a 2×2×3 factorial arrangement (incubator, breed and age), totaling 12 treatments with 6 replicates each. Each cart inside of an incubator was considered as an experimental unit that contained 5,040 eggs. Both single and multiple stage incubators contained 36 carts. The eggs were produced by Cobb 500 and Ross breeders at the beginning of the laying period (up to 30 weeks of age); between 40 and 45 weeks of age (middle age) and at the end of the laying period (>60 weeks of age).

The cloacal temperature was measured using an atrial thermometer. The measurements were taken at 3 randomly selected time points during (1) exit from the hatchery, (2) separation of chicks from debris, (3) in the dispatch room. The average temperature of the 10 chicks was calculated to obtain a single mean.

Each dependent variable was compared between the treatments using Analysis of Variance (ANOVA) and triple factor, followed by the Tukey's test. Once the samples were selected by means of a completely randomized design, a parametric analysis was performed with the assumption that the data were normal. Analyses were performed using SAS 9.O (Statistical Analysis System) and a level of significance was set at $p<0.05$.

RESULTS

There was a three-way interaction between age, breed and incubator type ($p<0.05$; Table 1) on the capacity of chicks to regulate body temperature at the three time points. Upon exit from the hatchery, the chicks had the closest means of thermal comfort (104-106°F) compared to the other time points.

Upon exit from the hatchery, chicks produced by young Ross breeders and those from old Cobb breeders that were incubated in a multiple stage incubator had greater difficulty

Table 1: Average body temperature of chicks at three time points

Breed	Incubator	Breeder age	Hatcher (°F)	Separate from debris (°F)	Dispatch room (T°)
Ross	ME	Y	102.6800	103.3700	100.88
		M	104.4200	103.5100	102.58
		O	104.4800	103.6700	103.36
	SS	Y	104.1700	103.5000	101.66
		M	105.0600	103.4400	103.42
		O	102.9300	102.0700	102.08
Cobb	ME	Y	103.6900	103.3700	102.81
		M	104.3700	105.6100	102.72
		O	103.1900	102.6200	102.10
	SS	Y	103.5200	103.4800	102.92
		M	104.2400	103.1100	102.00
		O	103.5400	103.7300	101.90
p-value			<0.00010	<0.0001	<0.0001
Breed			0.63970	0.0164	0.5677
Incubator			0.41560	0.0048	0.5796
Breeder age			<0.0001	0.0001	0.0073
Breed *incubator			0.6073	0.8025	0.2492
Breed *breeder age			0.01750	0.0779	<0.0001
Incubator *breeder age			0.44730	0.0021	0.0124
Breed* incub. *breeder age			0.00200	<0.0001	0.0041

ME: Multiple stage incubator, SS: Single stage incubator, Y: Young breeder, M: Middle old breeder, O: Older breeder

Table 2: Three-way interaction between breed, breeder age and incubator type on average body temperature of chicks

Factors	Interaction	Body temperature (°F)	p-value
Cobb	ME×Y	104.17^{A}	0.0013
Ross		102.68^{B}	
Ross	ME×O	104.48^{A}	0.0110
Cobb		102.93^{B}	
Y	ME×Cobb	104.17^{B}	0.0001
M		105.06^{A}	
O		102.93^{C}	
Y	ME×Ross	102.68^{B}	0.0018
M		104.42^{A}	
O		104.48^{A}	
SS	Ross×Y	103.69^{A}	0.0012
ME		102.68^{B}	

ME: Multiple stage incubator, SS: Single stage incubator, Y: Young breeder, M: Middle age breeder, O: Older breeder. A, B: Different capital letters in the columns indicate statistical difference by the Tukey test (p<0.05), The same letters indicate lack of significant differences

in maintaining body temperature, even when heat loss to the environment was lower and similar to that at the hatchery (Table 2).

Temperature trends upon separation of chicks from eggshell debris were similar to those seen upon exit from the hatchery, except that when eggs were incubated in a single-stage incubator, the chicks had a lower capacity to maintain body temperature, particularly those chicks from eggs produced by old and young Ross and Cobb breeders, respectively (Table 3). Colder temperatures when debris is separated from the chicks may result in this decreased body temperature. However, this possibility does not explain the critical low body temperature of some chicks, especially since

Table 3: Three-way influence of breed, breeder age and incubator type on chick body temperature following separation from eggshell debris

Factors	Interaction	Body temperature (°F)	p-value
Cobb	SS×O	103.72^{A}	0.0029
Ross		102.06^{B}	
Cobb	ME×M	105.61^{A}	0.0017
Ross		103.50^{B}	
Ross	ME×O	103.66^{A}	0.0241
Cobb		102.62^{B}	
Y	SS×Cobb	103.48^{B}	0.0364
M		103.11^{B}	
O		103.72^{A}	
Y	SS×Ross	103.50^{A}	0.0009
M		103.44^{A}	
O		102.06^{B}	
Y	ME×Cobb	103.37^{B}	<0.0001
M		105.61^{A}	
O		102.62^{B}	
Y	ME×Ross	102.68^{B}	<0.0001
M		104.42^{A}	
O		104.48^{A}	
ME	Cobb×M	105.61^{A}	0.0005
SS		103.11^{B}	
SS	Cobb×O	103.72^{A}	0.0024
ME		102.62^{B}	
ME	Ross×M	103.66^{A}	0.0104
SS		102.06^{B}	

ME: Multiple stage incubator, SS: Single stage incubator, Y: Young breeder, M: Middle old breeder, O: Older breeder. A, B: Different capital letters in the columns indicate statistical difference by the Tukey test (p<0.05), Equal letters not differ significantly between themselves

the increased manipulation at this stage should raise body temperature and again suggests that these chicks have deficient thermoregulation.

In the dispatch room, cold can be intensified by several factors, including exposure to spray vaccine, wet boxes and

Table 4: Interactions between breed, breeder age, incubator type and average body temperature of chicks in the dispatch room

Factors	Interaction	Body temperature (°F)	p-value
Cobb	SS×Y	102.92^{A}	0.0230
Ross		101.59^{B}	
Ross	SS×M	103.41^{A}	0.0029
Cobb		101.99^{B}	
Cobb	ME×Y	102.80^{A}	0.0036
Ross		100.87^{B}	
Ross	ME×O	103.35^{A}	0.0022
Cobb		102.09^{B}	
Y	SS×Ross	101.59^{B}	0.0005
M		103.41^{A}	
O		102.08^{B}	
Y	ME×Ross	100.87^{C}	<0.0001
M		102.57^{B}	
O		103.35^{A}	
ME	Cobb×M	102.72^{A}	0.0006
SS		101.99^{B}	
SS	Ross×Y	101.59^{A}	0.0466
ME		100.87^{B}	
ME	Ross×O	103.35^{A}	0.0625
SS		102.08^{B}	

A, B: Different capital letters in the columns indicate statistical difference by the Tukey test (p<0.05), Equal letters not differ significantly between themselves

ambient temperature, which can all present even greater challenges for thermal regulation capacity of chicks. Here we observed that the chicks did not reach the minimum comfort temperature (104-106°F; Table 4).

DISCUSSION

Environmental factors such as incubation temperature can induce permanent, epigenetic changes that affect bodily functions and responses to the environment[6]. Tzschentke and Tatge[6] reported that appropriate development of the thermoregulatory system of chicks during incubation can result in more efficient thermal regulation after hatching.

Furthermore, other studies showed that exposure of embryos to high[7] or low temperatures[8] during incubation (day 7-16) may improve the ability of chickens to cope with cold[9,10] or heat[11-14] stress.

The similar results seen for incubation at decreased or increased temperature can be related to differences among breed, breeder age and incubator type. To our knowledge, this is the first study to demonstrate the different ways that these factors relate to one another.

Compared with single-stage incubators, multiple stage incubators have variable heating at different embryonic stages[5] and cooling that is divided by gas exchange function[15]. We found that chicks produced using multiple stage incubators had less efficient temperature control during incubation relative to those produced with single-stage incubators.

This temperature control deficiency during incubation in a multiple stage incubator may reflect inadequate development of the thermoregulatory system in the birds (i.e., the Hypothalamic-Pituitary-Thyroid axis and the Hypothalamic-Pituitary-Adrenal axis) during embryonic development[16-18]. Indeed, we observed the lowest average temperature for chicks incubated in multiple stage incubators.

The size of chicks affects thermal regulation ability[19], which can also vary depending on breeder age or breed. Smaller chicks have high body surface to weight ratios that make them more sensitive to temperature loss mainly because body fat helps retain body heat[20,21]. Our finding that Ross chicks from young breeders were smaller and thus tended to be more sensitive to cold was consistent with these earlier studies.

Meanwhile, chicks produced by older Cobb breeders also exhibited effects associated with development of the thermoregulatory system, since, despite their larger size, they had lower quality. This outcome may be due to decreased calcium retention and pore diameter of eggshells produced by this breed, particularly with age[22], that render the shell less efficient in controlling embryo temperature during incubation. Furthermore, very large eggs may be inadequately cooled during incubation due to the different relationship between volume and surface area[23,24,25].

In addition to the factors examined here, genetic modifications selected for during poultry breeding can affect the ability of birds to respond to extreme environmental temperatures[26], which could explain the lower temperatures of the chicks in this study. However, deficiencies in maintaining warm body temperatures could have long-term benefits as an adaptation to heat stress[27]. Results from this study indicate that providing a suitable ambient temperature is important for early-stage chicks that are more sensitive to cold (both in the hatchery and the farm).

CONCLUSION

Chicks produced by both young Ross hens and old Cobb hens have a lower capacity to maintain body temperature when incubated in a multiple stage incubator and in the dispatch room. Therefore, higher ambient temperatures are needed for one-day old chicks from these types of hens.

SIGNIFICANCE STATEMENT

This study discovered that a specific breed resulted in chicks with lower capacity to remain warm (deficit in body temperature regulation) in the hatchery mainly when other factors are involved like old breeder and multiple stage incubator. This problem is bigger in the dispatch room because there is a lower environment temperature. This study will help the researcher to uncover the ways to improve the welfare of chicks. Thus a new theory on the factors related to the body regulation of chicks may be arrived at.

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