Brazilian Journal of Poultry Science
Revista Brasileira de Ciência Avícola

ISSN 1516-635X Oct - Dec 2016

Special Issue 2 Incubation / 017-026

http://dx.doi.org/10.1590/1806-9061-2015-0146

Effect of Incubator Type and Broiler Breeder Age on Hatchability and Chick Quality

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Keywords

Embryo, hatch window, multiple-stage and single-stage incubation.

Submitted: August/2015
Approved: September/2015

ABSTRACT

Two experiments were conducted to evaluate the effect of broiler breeder age and incubator type on hatching parameters, hatch window, embryo diagnosis results, and hatchling physical quality. The treatments consisted of a combination of three broiler breeder ages (29, 35 and 59 weeks of age) and two incubator types (single stage, SS; or and multiple stage, MS). A completely randomized design in a 3x2 factorial arrangement was applied. In Experiment I, 1,896 eggs were used and 360 eggs in Experiment II. There was an interaction between breeder age and incubator type only for hatchling physical quality score. Independently of incubator type, hatchability rate, late embryo mortality, and egg contamination were higher in the eggs laid by older breeders (59-wk-old). Early mortality (0-4 days) was higher in the embryos from young breeders (29-wk-old). A shorter hatch window birth was obtained in the SS incubator, resulting in higher hatchling body weight relative to egg weight, and better hatchling physical quality score. Both types of incubators provide good conditions for embryo development; however, the physical quality of chicks derived from eggs from intermediate-aged breeders (35-wk-old) is better when eggs are incubated in SS incubators.

INTRODUCTION

Incubation is an essential stage in broiler production (Tona *et al.,* 2003). According to Bramwel (2002), incubation yield is affected particularly by egg fertility, incubation conditions (incubators), and hatchery management. In addition of obtaining excellent hatchability, industrial hatcheries seek to obtain good chick quality (Willemsen *et al*., 2008). According to Ulmer-Franco *et al*. (2010), good quality hatchlings are active, weigh between 40 and 44 g, their navel is healed upon removal from the hatcher, and their down is dry. Heavier hatchlings are also heavier at slaughter age compared with light hatchlings (Stringhini *et al*., 2003). Molenaar *et al*. (2008) also reported that well-hydrated hatchlings with completely healed navels showed better performance during the rearing period (1-42 d).

Hatchling physical quality may be affected by breeder age. Chicks derived from large eggs have higher body weight and develop faster due to the higher availability of nutrients present in the egg compared with those from small eggs (Lourens *et al.,* 2006).Young breeders lay eggs with thicker eggshells than those of older breeders, and therefore, less oxygen is supplied to the embryos, which may explain their slower metabolism and development (Hamidu *et al*., 2007).

Hatchling physical quality is also influenced by temperature, humidity, and ventilation inside the incubator. Small deviations from the recommended values may increase embryonic mortality and consequently, reduce hatchability rates (Bolzan *et al.,* 2008). According to Calil (2007), the physiological needs of the embryo change according to their developmental stage and, therefore, the maintenance of temperature during incubation is one of the most important factors influencing embryonic development.

Currently, two types of incubators (multistage or single stage) are used in commercial hatcheries. Multistage incubators (MS) set two or three loads of eggs per week. Therefore, lots of eggs from different broiler breeder farms and embryos at different stages of development are incubated in a single machine (Moro, 2007). When eggs are incubated in MS machines, older embryos transfer heat to younger embryos, establishing a thermal balance within the machine; however, they may also cause excessive increase of the temperature inside the setter, triggering embryo mortality (Gonzales *et al.*, 2009).

On the other hand, single-stage incubators (SS) are fully loaded with a single egg lot. Therefore, all embryos have the same developmental stage, allowing temperature, humidity, and ventilation to be set according to the embryos' needs (Molenaar *et al.,* 2010). SS machines are recommended for the incubation of eggs of modern high-yield broiler strains, which embryos generate more heat than the slow-growing strains (Boerjan, 2004).

In a study with eggs laid by a flock of breeders with the same age, an improvement in the incubation parameters was observed when eggs were incubated in a SS machine compared with a MS machine (Calil, 2013). However, some hatcheries are not able to obtain a sufficient number of eggs laid by a single breeder flock to fully load an incubator, and consequently eggs from flocks of breeders of different ages are set in a single machine.

This study aimed at determining the best type of machine to incubate eggs from broiler breeders of different ages by analyzing hatching parameters and hatchling physical quality.

MATERIALS AND METHODS

All procedures used in the experiments were approved by the Ethics Committee on Animal Use from the Federal University of Goiás, Brazil.

In total, 2,256 hatchable eggs laid by a commercial flock of Cobb broiler breeders were evaluated in two experiments. Eggs were obtained from three broiler breeder flocks, with 29 (young breeders), 35 (intermediate-age breeders), and 59 (old breeders) weeks of age (wk). Eggs were stored at 16°C and 75% RH for three days, and warmed to room temperature (22°C) before setting. The eggs of 29-wk-old, 35-wk-old, and 59-wk-old broiler breeders weighed 59.16 g ± 1.25, 60.02 g ± 1.01, and 69.70g ± 1.09, respectively.

In Experiment I, a total of 1,896 eggs laid by COBB® commercial broiler breeders of the above-mentioned ages were used. Eggs were distributed according a randomized block experimental design in a 2 x 3 factorial arrangement, consisting of two types of incubation machines (SS or MS) and three breeder ages (29, 35, or 59 weeks), totaling six treatments with four replicates of 79 egg trays each. Blocks corresponded to the position of the tray (upper, middle, or lower position) in the trolley.

The multistage incubator (CASP® CMg 125 HT) had a capacity of 124,416 eggs, distributed in 36 trolleys with 36 trays each. Tray capacity was 96 eggs. The single stage incubator (CAPS® Ug 62 HT) had a capacity of 61,920 eggs, distributed in 12 trolleys with 60 trays each. Tray capacity was 86 eggs. In Experiment I, eggs were set in three trolleys with three trays each in the MS machine, and in three trolleys with four trays each in the SS machine. In the Experiment II, eggs were set in two trays of one trolley per incubator type. Non-experimental eggs were set to complete the full capacity of both machines.

The SS machine was equipped with infrared sensors that constantly monitored eggshell temperature, coupled with the setter control to provide the ideal temperature for each embryo developmental stage, as well as an integrated weighing system and CO_2 level control. In both incubators, initial temperature was set at 37.4 (98.9ºF) and relative humidity at 58%. Temperature and relative humidity were automatically set after the beginning of the incubation in the SS machine.

On day 18 of incubation (432 h), *in-ovo* vaccination against Marek's disease was performed. Eggs were then transferred to a hatcher (CASP® 108 HR), with a capacity of 19,264 eggs, set to maintain 37.3ºC (68.26ºF) temperature and 42.0% RH. The experiment was completed at 504 hours of incubation, when hatched chicks were removed from the hatch baskets.

The evaluated parameters were hatchability percentage relative to the number of incubated eggs and relative to the number of fertile eggs. The percentage of fertile eggs was determined by a fertility test by carried out by the hatchery personnel for each of the three broiler-breeder age groups, as 92%, 91%, and 85% for 29-, 35-, and 59-wk-old breeders, respectively. Hatching was monitored from 468 hours to 504 hours of incubation. Hatch window comprised the period between the first and the last chick hatched in each basket (468-504 h of incubation). The hatcher was opened every six hours to count the number of hatched chicks, after which, the basket was returned to the hatcher.

At the end of the hatch window, the unhatched eggs were submitted to embryo diagnosis, considering four stages of embryonic mortality: phase I included the period between zero to four days of embryonic development; phase II, between five to ten days of embryonic development; phase III, 11 to 18 days of embryonic development; and phase IV, between 19 and 21 days of embryonic development. The number of pipped eggs with dead and live chicks, eggs with dead and live chicks presenting any abnormalities, live and not pipped, and eggs with evidence of bacterial or fungal contamination were counted.

The incubation parameters were subjected to analysis of variance and means were compared by Tukey's test ($p<0.05$). The embryo diagnosis data were submitted to Fisher's exact test ($p<0.05$).

In Experiment II, 360 fertile eggs were used to evaluate the effect of incubator type and breeder age on hatchling physical quality. Therefore, a completely randomized experimental design in a 2x3 factorial arrangement (incubator type x broiler breeder age), totaling six treatments, with six replicates of one egg each, was applied. After selection, eggs were individually weighed on a precision scale (0.01g) to obtain initial egg weight. Incubation management was identical to that applied in Experiment I. On day 18 of incubation (432 hours), the trolleys were removed from the setter and 360 eggs were weighed and transferred to the vaccination room. Eggs were then individually placed in the hatch baskets, fitted with air-permeable metal divisions to allow identifying the hatchlings and their respective eggshells. The experiment was completed at 504 hours of incubation, when hatched chicks were removed from the hatch baskets.

The evaluated parameters were egg weight loss during incubation, body weight of the hatchlings with dried down, chick weight at time of dispatch, chick weight loss between hatch and dispatch, hatchling weight relative to egg weight at setting, chick weight at dispatch relative to egg weight at setting, yolk-free chick weight (net weight), residual yolk weight relative to chick weight at dispatch, and net weight relative to chick weight at dispatch. In order to determine residual yolk weight, hatchlings were euthanized by cervical dislocation, and the residual yolk was removed and weighed. Hatchling physical quality was evaluated using the method proposed by Tona *et al*.(2003) and hatchling length was determined according to Wolanski *et al*. (2007).

Quantitative data were submitted to analysis of variance and means were compared by Tukey's test at 5% probability level ($p<0.05$). Qualitative data (hatchling physical quality score) were subjected to analysis of variance and the means compared by the test of Kruskal-Wallis, at 5% probability level ($p<0.05$). All analyses were performed using the R software (2010).

RESULTS AND DISCUSSION

No interaction between incubator type and breeder age ($p>0.05$) was observed for hatchability or hatchability/fertile eggs (Table 1). This indicates that both incubators were able to meet the temperature, humidity, and ventilation requirements of the embryos of breeders of the different evaluated ages. Hatchability percentage was not different ($p>0.05$) between incubator types, independently of breeder age. These results differ from the findings of Bennet (2010), who obtained higher total hatchability with an SS machine (86.60%) compared with a MS machine (84.01%), but with eggs laid by breeders of the same age. Molenaar *et al*. (2010) also mentioned that SS incubation systems promote higher hatchability than MS systems. However, in the present experiment, no hatchability differences were observed between SS and MS incubators. The fact that eggs from breeders of different ages were set in a same machine may have affected the results, because it was not possible to adjust SS incubator temperature and humidity settings according to egg size. The SS system is more appropriate to incubate eggs from the same broiler breeder age.

Eggs from 59-wk-old breeders presented lower hatchability ($p<0.05$) compared with the two other evaluated breeder ages. These results are consistent with the findings of Rosa *et al*. (2002), who found lower hatchability of the eggs of 62-wk-old breeders (82.80%) than of 34-wk-old breeders (86.60%). According to Almeida *et al*. (2006), the lower hatchability of eggs laid by older breeders may be explained by the decline in eggshell quality as breeders age, resulting in higher embryo mortality. The eggs of older breeders have thinner eggshells with the higher

Table 1 – Hatchability of the eggs laid by broiler breeders of different ages and incubated in two different types of incubator (Experiment I)

Breeder age (weeks)	Hatchability (%)	Hatched/fertile eggs (%)	Hatch window[1] (h)
29	89.81^{a} ± 0.31	92.32^{a} ± 0.20	24.00 ± 0.23
35	88.37^{a} ± 0.16	92.96^{a} ± 0.11	19.00 ± 0.35
59	81.04^{b} ± 0.79	88.26^{b} ± 0.55	23.00 ± 0.23
Incubator type			
Single stage	87.38 ± 0.30	92.51 ± 0.25	19.50^{b} ± 0.09
Multistage	84.34 ± 0.24	89.45 ± 0.21	25.00^{a} ± 0.18
p-value			
Breeder age	0.0021	0.0212	0.1091
Incubator type	0.3712	0.2507	0.0028
Age *vs.* incubator type	0.3602	0.4005	0.8812

$^{a-b}$ Means within a column with no common superscript differ significantly by Tukey's test (p<0.05).

[1]Mean hatch window: period (h) between the first and the last chick that hatched in the basket.

number of pores relative to younger breeders, allowing gas exchange between the egg and the environment, and therefore, higher moisture is required during incubation to prevent excessive egg dehydration (Rosa *et al.*, 2002). Egg dehydration has a decisive role during incubation, as it affects embryonic development and hatchability rates (Tona *et al.* 2001).

Hatch window (interval between the first egg and the last egg to hatch, in hours) were not influenced by breeder age (p>0.05). On the other hand, Decuypere & Bruggeman found that breeder age of affected hatch window.

Hatch window (Table 1) was affected by incubator type (p<0.05), and the SS machine promoted a shorter hatch window (Figure 1). It was observed that the chicks incubated in the MS incubator were still hatching at the end of the experiment (504 hours of incubation), where as hatching in the SS incubator was completed at 497 hours of incubation. According to Wineland *et al*. (2008), MS incubators are not able to supply the temperature requirements of all developing embryos, as such requirements vary with embryo developmental age.

Hatching at the end of hatch window is a disadvantage, because the time these chicks remain in hatcher is not sufficient to allow complete navel healing. In addition, they are sent to the chick processing room with their down still wet, and are consequently classified as unfit. Boerjan (2004) reported that the hatch window influences broiler performance in the field. Chicks that hatch at the beginning or end of the hatch window have lower growth potential during the first week of rearing than those that hatch in the intermediate period (Willemse *et al.,* 2008). In commercial hatcheries, hatch windows are typically of 24 to 48 hours. Chicks that hatch early remain a long time inside the hatcher, suffering water and food deprivation (Calil, 2013), which may result in low weight gain, activation of the immune system, reduced expression of digestive enzymes and slower organ development (Willemse *et al*., 2008).

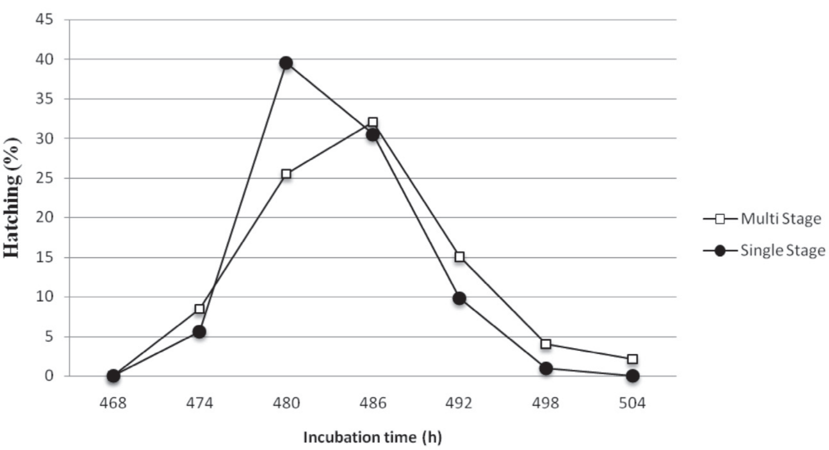

Figure 1 – Hatch window of eggs laid by broiler breeder of different ages (29, 35 and 59 weeks) according to incubator type (Experiment 1).

In this experiment, the eggs laid by breeders of different ages were incubated in a same SS machine, which may have favored the embryonic development of eggs laid by intermediate-aged breeders, possibly because the temperature and relative humidity of the SS machine met the requirements of the embryos of medium-sized eggs laid by the 35-wk-old broiler breeders.

Table 2 shows the embryo diagnosis results according to incubator type. Embryo mortality was not influenced (p>0.05) by incubator type. These results are consistent with the findings of Mauldin *et al*. (2006), who did not find any influence of SS or

MS incubators on early, intermediate, and late embryo mortality, and concluded that both systems offered similar incubation conditions for embryo development.

Table 2 – Embryo diagnosis results (%) of unhatched eggs incubated in single-stage or multistage incubators (Experiment I)

	Incubator type		
Diagnosis (%)	Single-stage	Multistage	*p*-value
Infertile eggs	3.00	3.19	0.8946
MI	5.67	4.16	0.1342
MII	1.04	1.35	0.6742
MIII	1.73	1.64	0.9100
MIV	1.04	0.48	0.1800
Pipped, dead	0.11	0.48	0.2290
Pipped, alive	0.46	0.87	0.4039
Abnormal, alive	0.00	0.00	1.0000
Abnormal, dead	0.00	0.09	1.0000
Non-pipped, alive	0.00	0.19	0.5016
Contaminated eggs	1.73	0.87	0.1024

MI (mortality from 0 to 4 days of embryo development); MII (mortality from 5 to 10 days of embryo development); MIII (mortality from 11 to 17 days of embryo development); MIV (mortality from 18 to 21 days of embryo development).

Table 3 shows the embryo diagnosis results as a function of breeder age. Breeder age did not affect ($p>0.05$) embryo mortality from five to ten days (MII) or from 11 to 17 days (MIII) of incubation, or the number of pipped and alive, pipped and dead, abnormal and alive, abnormal and dead, and non-pipped alive chicks. However, significant differences ($p<0.05$) in infertile egg, embryo mortality from zero to four days and from 18 to 21 days of incubation, and percentage of contaminated eggs were observed as a function of breeder age. Eggs from 29-wk-old breeders presented lower fertility rate ($p<0.05$) compared with those from 59-wk-old breeders, but were not different from those laid by 35-wk-old breeders. These results support the theory Meijerhof *et al*. (1991) and Fasenko *et al*. (1995), who stated that fertility is inversely proportional to breeder age.

Embryos from 59-wk-old breeders presented higher early mortality ($p<0.05$). Tanure *et al*. (2009) reported older breeder eggs present worse eggshell quality compared with younger breeders, and lose more water when relative humidity inside the incubator is low, which explains the higher early embryo mortality.

Higher ($p<0.05$) late embryo mortality (18-21 days of incubation) was observed in the eggs laid by 29-wk-old breeders compared with the other age groups. These results agree with Francisco (2012),

Table 3 – Embryo diagnosis (%) of unhatched eggs according to broiler breeder age relative to the total number of incubated eggs (Experiment I)

	Breeder age (weeks)			
Diagnosis (%)	29	35	59	*p*-value
Infertile eggs	1.46^{b}	2.93^{b}	6.84^{a}	≤0.0001
MI	2.38^{c}	3.29^{b}	5.47^{a}	0.0117
MII	1.09	1.64	0.99	0.5961
MIII	1.83	1.64	1.61	0.9737
MIV	1.46^{a}	0.36^{b}	0.49^{b}	0.0500
Pipped, dead	0.36	0.36	0.24	1.0000
Pipped, alive	1.09	0.54	0.49	0.4089
Abnormal, alive	0.00	0.00	0.00	1.0000
Abnormal, dead	0.00	0.19	0.00	1.0000
Non-pipped, alive	0.00	0.00	2.23	1.0000
Contaminated eggs	0.54^{b}	0.54^{b}	2.23^{a}	0.0062

$^{a-c}$ Means within a line with no common superscript differ significantly according to Fisher's exact test ($p<0.05$).

MI (mortality from 0 to 4 days of embryo development); MII (mortality from 5 to 10 days of embryo development); MIII (mortality from 11 to 17 days of embryo development); MIV (mortality from 18 to 21 days of embryo development).

who observed higher embryo mortality on days 18 to 21 of incubation in the eggs from 33-wk-old breeders compared with those from 61-wk-old breeders. The high late mortality of embryos from young breeders may be explained by oxygen deficiency. According to Vieira & Pophal (2000), broiler breeders in the beginning of lay produce thicker albumen, which may slow down gas exchange and hinder yolk sac absorption, resulting in deficient supply of nutrients to the embryo. Around 90% of the energy produced by the embryo derives from the oxidation of the fatty acids present in the yolk. Therefore, oxygen deficiency at the end of incubation hinders fatty acid oxidation, slowing embryo development, which results in high late embryo mortality (Lara *et al*., 2005).

A higher number of contaminated eggs was observed in the group of 59-wk-old breeders, as determined by the presence of gas and foamy egg content, compared with 29 and 35-wk-old breeders. In general, older breeders lay larger eggs, which reduced density and higher porosity of the eggshell favor the entry of bacteria and fungal spores (Silva *et al.,* 2003).

The results of the incubation parameters evaluated in Experiment II are shown in Table 4. There was no interaction between breeder age and incubator type for any of the studied parameters ($p>0.05$). Incubated egg weight was influenced by breeder age ($p<0.05$), with heavier eggs laid by older breeders. Breeder age also affected egg weight loss (p<0.05), with greater weight loss between incubation and transfer to the hatcher observed in the eggs of older breeders (59 weeks of age). These results are consistent with the findings of Santos *et al*. (2009) and Ulmer-Franco *et al*. (2010), who attributed this higher egg weight loss to the higher eggshell conductance of older-breeder eggs.

Lower egg weight loss during incubation was obtained with the SS machine compared with the MS machine (p<0.05), independently of breeder age; however, the egg weight loss obtained with both machines were within acceptable values. According to Gonzales & Mello (2012), egg weight loss of 10 to 14% up to transfer to the hatcher indicates adequate gas exchange, and consequently, good hatchling physical quality. However, Morita *et al*. (2010) suggested that young breeder eggs should not be incubated in MS machines, because temperature may be low at the beginning of incubation, resulting in insufficient egg weight loss, which may impair embryo development.

The results of incubated egg weight, egg weight loss, hatchling weight, hatchling weight loss between hatch and dispatch, and hatchling weight relative to egg weight are shown in Table 4. There was no interaction between incubator types and breeder age for any of the parameters studied (p>0.05)

Hatchling weight at hatch and hatchling weight at dispatch were influenced both by breeder age and incubator type (p<0.05). Chicks from 59-wk-old breeders were heavier compared with those from 29- and 35-wk-old breeders. Lara *et al.* (2005), Gonzales & Mello (2012), Oviedo-Rondon *et al.* (2009) also reported that hatchling weight is correlated with incubated egg weight.

Chicks from eggs incubated in SS machine were heavier at hatch (p<0.05) than those incubated in the MS machine, as well as heavier at dispatch (p<0.05). This indicates that the eggs that lost less weight during incubation (p<0.05) produced heavier chicks, which may be associated with lower dehydration of the embryo during incubation. According to Gonzales & Mello (2012), the water present in the yolk is partially accumulated in the embryo tissues and in the yolk sac. However, Wineland *et al*.(2008) did not observe any differences in chick weight in a study comparing SS with MS incubators in a commercial hatchery. Oviedo-Rondón *et al*.(2009) found that chicks incubated in SS machines gained more weight during rearing up to 56 days of age.

Chicks from 29-wk-old breeders presented higher body weight values relative to incubated egg weight (p<0.05), indicating that embryos from smaller eggs utilize the egg content better than those from larger eggs. Regarding incubator type, eggs incubated in the SS machine yielded higher hatchling weight relative to incubated egg weight (p<0.05).

Hatchling weight loss between the hatcher and dispatch was not influenced by breeder age (p>0.05). These results are consistent with the findings of Almeida *et al*. (2006). On the other hand, hatchling weight loss between the hatcher and dispatch was affected (p<0.05) by incubator type. Chicks incubated in the SS machine lost more weight between the

Table 4 – Incubation parameters as a function of breeder age and incubator type (single-stage or multistage) (Experiment II)

Egg weight and chick weight							
Breeder age (weeks)	Egg weight[A] (g)	Egg weight loss (%)	Hatchling weight (g)	Hatchling weight relative to egg weight (%)	Chick weight at dispatch (g)	Low chick weight at dispatch (g)	Low chick weight at dispatch (%)
29	59.20[b] ± 0.23	10.33[b] ± 0.31	44.19[b] ± 0.24	74.63[a] ± 0.56	42.3[b] ± 0.48	1.89 ± 0.38	4.27 ± 0.36
35	59.92[b] ± 0.34	11.62[b] ± 0.21	43.88[b] ± 0.23	73.25[b] ± 0.66	42.1[b] ± 0.45	1.78 ± 0.29	4.06 ± 0.26
59	69.80[a] ± 0.79	12.09[a] ± 0.12	50.97[a] ± 0.26	72.82[b] ± 0.78	49.01[a] ± 0.40	1.96 ± 0.43	3.82 ± 0.41
Incubator type							
Single-stage	62.04 ± 0.81	10.95[b] ± 0.70	47.27[a] ± 0.19	73.79[a] ± 0.12	45.53[a] ± 0.14	2.07[a] ± 0.19	4.55[a] ± 0.18
Multistage	63.01 ± 0.69	11.87[a] ± 0.73	45.61[b] ± 0.13	73.28[b] ± 0.20	43.53[b] ± 0.19	1.73[b] ± 0.78	3.66[b] ± 0.71
p-value							
Breeder age	≤ 0.001	≤ 0.001	≤ 0.001	≤ 0.001	≤ 0.001	0.5345	0.3523
Incubator type	0.1323	≤ 0.001	≤ 0.001	0.0329	≤ 0.002	0.0049	0.0018
Age *vs.* incubator	0.2219	0.1027	0.6789	0.8117	0.4445	0.5901	0.3443

[a–b] Means within a column with no common superscript differ significantly according to Tukey's test (p<0.05).

[A] Egg weights are represented as mean ± SEM of all eggs set for incubation.

hatcher and dispatch than those incubated in the MS machine. These results may be explained by the shorter hatch window obtained with the SS machine, as the hatched chicks remained about six hours in the hatcher with no access to water or food, consequently consuming the residual yolk. It is recommended that the hatchery that used the single-stage machine should accompany the hatch window and anticipate the removal of chicks to prevent excessive dehydration of the hatchlings.

The results of the yolk sac weight, yolk sac weight relative to chick weight at dispatch, yolk-free chick weight, chick length (cm), and hatchling physical quality score are shown in Table 5.There was an interaction between the incubator type and breeder age only for physical quality score ($p<0.05$) (Table 6), but not for the other physical quality parameters ($p>0.05$).

Yolk sac weight was influenced ($p<0.05$) by breeder age, with the heaviest yolk sacs obtained in chicks from 59-wk-old breeders, and the lightest in those from 35-wk-old breeders ($p<0.05$). These results agree with the findings of Joseph & Moran Jr. (2005) and Riccardi *et al*. (2009), who determined higher yolk sac weight in hatchlings from older hens. The higher residual yolk weight of the chicks from older breeders is due to the higher volume of lipoproteins and minerals deposited in the yolk. Chicks from 35-wk-old breeders presented had lower yolk sac weight ($p<0.05$), which may be related to greater absorption of the yolk sac, that is, better nutrient utilization by these birds. This result is consistent with the better physical quality scores of the chicks from those breeders (Table 5).

Yolk-free chick weight was influenced by breeder age ($p<0.05$); the chicks from 59-wk-old breeders were heavier compared with the others. However, Nangsuay *et al*. (2013), evaluating breeders of different ages (29 and 53 weeks) and similar egg weights, did not find any differences in yolk-free chick weight. The yolk-free chick weight results obtained in the present study support the theory of Sklan *et al*. (2003) that heavier eggs produce heavier chicks. Chick live and yolk-free chick weights are directly related to the amount of nutrients available to the embryo.

Chicks from 59-wk-old breeders were longer compared with those from 29- and 35-wk-old breeders. According to Molenaar *et al*. (2008), hatchling length is one of the best indicators of early broiler performance. This theory was confirmed by Van der Pol *et al.* (2013), who found a positive correlation between hatchling body length and broiler performance in the starter phase. Incubator type also affected chick length, with higher values obtained for the SS incubator ($p<0.05$) compared with the MS machine.

Table 6 presents the effect of the interaction between incubator type and breeder age on hatchling physical quality score. Chicks incubated in the SS machine presented better physical quality score, independently of breeder age. However, the quality of chicks from 35-wk-old breeders incubated in SS machines was better quality compared with those from 29- and 59-wk-old breeders. These results partially corroborate the findings of Bennet (2008), who reported that SS incubators produce more active, better hydrated, and more viable chicks than MS machines. The better physical quality score of hatchlings incubated in the SS machine may also be attributed to better hydration, considering that in the present experiment, SS-incubated chicks lost less weight between the hatcher and dispatch, and hatched during a shorter hatch window.

Table 5 – Hatchling physical quality as a function of breeder age and incubator type (Experiment II)

Hatchling physical quality					
Breeder age (weeks)	Yolk sac weight (g)	Yolk sac weight(%)	Yolk-free chick weight (g)	Chick length (cm)	Chick quality score[1]
29	$4.37^b \pm 0.24$	$10.61^a \pm 0.74$	$35.77^b \pm 0.56$	$17.66^c \pm 0.10$	91.18 ± 0.93
35	$3.52^c \pm 0.31$	$8.64^b \pm 0.68$	$37.17^b \pm 0.48$	$17.86^b \pm 0.07$	95.30 ± 0.87
59	$5.06^a \pm 0.28$	$10.55^a \pm 0.83$	$42.80^a \pm 0.40$	$18.32^a \pm 0.15$	92.28 ± 1.01
Incubator type					
Singlestage	4.48 ± 0.34	10.06 ± 0.76	$39.87^a \pm 0.44$	$18.22^a \pm 0.19$	95.47 ± 0.81
Multistage	4.17 ± 0.40	9.93 ± 0.68	$37.72^b \pm 0.53$	$17.61^b \pm 0.23$	91.06 ± 0.71
p-value					
Breeder age	≤ 0.0001	≤ 0.0001	≤ 0.0001	≤ 0.0001	0.0222†
Incubator type	0.6114	0.7698	≤ 0.0001	≤ 0.0001	0.0401†
Age*vs.* incubator	0.2627	0.2731	0.6613	0.2144	0.0412†

[a–c] Means within a column with no common superscript differ significantly according to Tukey's test ($p<0.05$). † Means differ significantly according to Kruskal-Wallis test ($p<0.05$).

[1] Score according to Tona *et al*. (2003).

Table 6 – Effect of the interaction between broiler breeder age and incubator type on hatchling physical quality score.

Hatchling physical quality score (0-100 points)			
Incubator type	Breeder age (weeks)		
	29	35	59
Singlestage	93.14Ab	96.12Aa	92.91Ab
Multistage	91.80Ba	92.12Ba	90.01Ba

Values followed by different uppercase letters (column) and lowercase letters (line) are different by the Kruskal-Wallis test (p< 0.05).

Considering the better quality of the chicks from 35-wk-old breeders and incubated in the SS machine obtained in the present experiment, Tona *et al*. (2008), in a study on the quality of chicks derived from breeders of different ages and incubated in single-stage machine, also observed better physical quality of chicks from 35-wk-old breeders compared with those from 45-wk-old breeders.

CONCLUSIONS

Both single stage and multistage incubators provided adequate conditions for embryonic development, and did not affect the hatchability of eggs laid by breeders of different ages were incubated together. The SS incubator shortened hatch window and yielded better hatchling quality. When eggs laid by young, intermediate-aged and older broiler breeders were incubated same SS incubator, hatchlings derived from eggs laid by intermediate-aged breeders presented better physical quality score.

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