



Is the Eggshell Quality Influenced by the Egg Weight or the Breeder Age?

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ABSTRACT

For a long time, it has been said that eggshell quality decreases as eggs increase in size, but this increase is seen as the breeder age advances. Therefore, the present study aimed to evaluate the age and egg weight effect on Cobb 500 broiler breeders on eggshell quality. The quality measures tested were eggshell and mineral matter percentage, eggshell surface, shape and eggshell index, eggshell and membrane thickness and specific gravity. Eggs from 27, 31, 36, 40, 46, 48, 53, 58 and 63 week-old breeders were classified as eggs with a constant weight ranging from 63.8 ± 0.2 ; and eggs with increasing weight, according to age, being: 55.8g; 60g; 62.2g; 64.2g; 65.2g, 66.6g, 68.1g, 69.9g and 72.5g, respectively. The eggs weight did not influence the eggshell percentage and the specific gravity in seven of the nine tested ages, while for mineral matter in six of the nine studied ages. The shape and shell index, shell and membrane thickness are characteristics independent of egg weight. As the Cobb 500 broiler breeder age advanced, the mineral matter concentration and specific gravity maintained constant, the membrane thickness ($R2 > 70$) reduced, and other studied variables were influenced in the same way for both treatments. Thus, the eggshell quality decrease with the advancing age cannot be attributed solely to the increase of egg weight with consequent increase in the eggshell volume and the surface, but there are also age-related factors.

INTRODUCTION

The egg shell is a complex structure and represents the mineral reserve for embryo use (Solomon, 2010) as well as the physical protection for its growth (Ulmer-Franco *et al.*, 2010). Thus, good quality eggshells must occur throughout all the reproductive life of breeders since they influence the embryonic development (Tůmová & Gous, 2012). The eggshell quality can be affected by genotype, breeder age, room temperature (Tůmová *et al.*, 2014) and nutrition (Londero *et al.*, 2015; Tůmová & Gous, 2012), on the other hand, the laying time does not influence the eggshell quality (Gumulka *et al.*, 2010; Tůmová *et al.*, 2014).

The breeder age increasing results in egg production decrease and egg weight increase, which can be attributed to a decrease in the posture sequence (Rutz *et al.*, 2005) and to the increase in the breeder's weight (Tůmová & Gous, 2012). The eggshell quality deteriorates with the advancing of age (Machal & Simeonová, 2002), which is presumably related to the increase in egg size and consequent eggshell surface area (Tůmová & Ledvinka, 2009).

The eggs' weight from a flock of breeders of the same lineage and age shows distribution in the normal curve and can be influenced by



feeding time (Londero *et al.*, 2015), room temperature and laying time (Tůmová & Gous, 2012). Eggs laid at the beginning of the day, up to 7 a.m., are heavier (Zakaria & Omar, 2013). According to Iqbal *et al.* (2017), the egg weight influences the eggshell percentage and thickness, shape index and specific gravity which reduce with the weight increase.

A long time ago, it was established the concept that the eggshell quality decreases as a consequence of the egg weight increase and the maintenance of the eggshell amount deposited throughout the reproductive life of the breeder. However, there is a lack of research evaluating, together, the effects of the breeders age and egg weight on the eggshell quality. However, it is hypothesized that the decrease in the eggshell quality with the advancing age is due to the breeder "geriatric" matters and not only to the egg weight increase. Therefore, this study aimed to evaluate the geriatric effect in Cobb 500 broiler breeders in eggshell quality.

MATERIAL AND METHODS

This study was performed with Cobb 500 broiler breeders, which were housed in nine sites at the same breeding farm, of a Brazilian agroindustrial company that produces broiler chickens, reared in an integration system. The flocks were housed with an average 4-week following, according to the company's housing schedule and the studied ages were: 27, 31, 36, 40, 46, 48, 53, 58 and 63 weeks old. The breeders and eggs management followed the company's routine and regular activities. At the reproduction stage, females between 27 and 46 weeks of age consumed first laying ration (2850 kcal kg⁻¹, 16.4% CP and 3.27% Ca) and females between 47 and 63 weeks of age second laying ration (2800 kcal kg⁻¹, 15.5% CP and 3.45% Ca), being the rations produced in the same feed mill and using the same raw materials (Table 1). The daily egg production was sent to the hatchery in the late afternoon and stored under a temperature of 19°C and humidity 70%.

The experiment started in the hatchery where 600 hatching eggs from each flock, from the second and third laying collection, were

individually weighed in an accuracy scale of 0.01 g, to determine the mean weight (Ewt) for each studied age. From these eggs, two treatments were defined: T1) eggs with constant weight: 63.8 ± 0.2g, being this weight determined to occur at all ages and the lowest weight found at 63 weeks; and T2) eggs with increasing weight according to the breeders' age: being these the average weight found for each age with variation of ± 0,2g: 55,8g; 60g; 62.2g; 64.2g; 65.2 g, 66.6 g, 68.1 g, 69.9 g and 72.5 g. For each treatment, 15 eggs were separated within each age (135), totalizing 270 eggs. The experiment consisted of a factorial design in subdivided plot, with nine ages in the plot and two categories of egg weight in the subplot.

The eggs specific gravity of each experimental unit was determined by the method of immersion in saline solutions at concentrations of 1.065; 1.070; 1,075 and 1,080 (Hamilton, 1982). The eggs remaining at the bottom of the solution of 1.080 were considered as density 1.085.

The width and height of each egg were measured (Breadth-B; Length-L) with a digital caliper with an accuracy of 0.0001mm. From the weight and these measurements, the following parameters were calculated:

$$\text{Shape Index (Carter, 1968): } (B / L) \times 100$$

$$\text{Egg volume (Ayupov, 1976): } \text{Evol (cm}^3\text{)} = 0.523 \times L \times B^2$$

$$\text{Shell (Surface) area (Mueller \& Scott, 1940): } \text{SA (cm}^2\text{)} = 4.67 (\text{Ewt})^{2/3}$$

The eggshell was washed in running water for the removal of the adhered albumen and dried at room temperature. The eggshell absolute weight (Shell wt) was obtained in a 0.001g precision scale. From these measurements, the following parameters were calculated:

$$\text{Shell (\%)} = (\text{Shell wt} / \text{Ewt}) \times 100$$

$$\text{Shell index (Sauveur, 1988): } \text{SI (g/cm}^2\text{)} = (\text{Shell wt} / \text{Shell surface area}) \times 100$$

Table 1 – Average feed intake (g), energy intake (kcal), crude protein (CP) and calcium (Ca) of Cobb 500 broiler breeders.

	Age	27	31	36	40	46	48	53	58	63
	g/hen	165	165	163	160	157	152	150	148	146
Feedintake / day	kcal	470	470	465	456	447	433	420	414	409
	CP	27.1	27.1	26.7	26.2	25.7	24.9	23.3	22.9	22.6
	Ca	5.4	5.4	5.3	5.2	5.1	5.0	5.2	5.1	5.0



The eggshell with membrane thickness (EWMT) was obtained by a “conical tip” micrometer with an accuracy of 0.001 mm and performed in three regions of the meridian. Then, the eggshells were submerged in 10% sodium hydroxide solution (NaOH) for 10 minutes, washed in distilled water and dried at room temperature to measure the eggshell without membrane thickness (EWOMT). The eggshell membrane thickness was obtained by the difference:

$$\text{Eggshell membranes thickness EMT} = (\text{EWMT} - \text{EWOMT})$$

Afterwards, the eggshells were ground to determine the mineral matter content (MM) by incinerating the sample in a muffle oven at 500° C for 4 hours.

$$\text{MM}\% = (\text{final weight} / \text{initial weight} \times 100)$$

The obtained data were analyzed for the residue’s normality (Kolmogorov-Smirnov test; $p < 0.01$) and variances homogeneity (Levene test; $p < 0.01$). Afterwards, they were submitted to ANOVA (“Snedecor” F test, $p < 0.05$). The comparison between the egg types was performed by the Tukey test ($p < 0.05$) and regression models were used to monitor the effect of breeders’ age on eggs ($p < 0.05$; $R^2 \geq 70\%$).

RESULTS

The constant (T1) and increasing (T2) egg weight and the eggshells variables are presented in table 2. The eggshell percentage was not dependent of egg weight, exception at 27 and 63 weeks of age, when eggs with higher weight presented lower eggshell percentage.

Table 2 – Percentage of eggshell, shell weight, mineral matter, specific gravity, shape index, egg volume, eggshell area and shell index of constant (T1) and increasing (T2) weight eggs from Cobb 500 broiler breeders.

Age	Shell Percentage		Mineral Matter (%)		Specific gravity (g mL ⁻¹ of H ₂ O)		Egg volume (cm ³)		Shell area (cm ²)	
	T1	T2	T1	T2	T1	T2	T1	T2	T1	T2
27	9.85b	10.53a	96.88a	96.78a	1084a	1085a	58.46a	51.77b	74.59a	68.31b
31	10.22a	10.35a	96.74b	97.50a	1077a	1077a	59.34a	56.48b	74.61a	71.66b
36	10.22a	10.42a	95.87b	97.11a	1076a	1075a	59.17a	57.71b	74.59a	73.44b
40	9.77a	9.99a	97.28a	97.27a	1074a	1074a	59.67a	58.89b	74.63b	74.95a
46	10.43a	10.32a	97.33a	95.77b	1075a	1074a	59.57b	60.73a	74.75b	75.80a
48	9.95a	10.32a	97.15a	96.81a	1074b	1076a	59.10b	62.10a	74.64b	76.87a
53	10.46a	10.36a	97.29a	96.62a	1075a	1076a	59.11b	62.93a	74.65b	78.02a
58	10.66a	10.72a	96.77a	96.94a	1075a	1076a	58.86b	64.46a	74.63b	79.33a
63	10.52a	9.50b	96.92a	97.44a	1077a	1074b	59.20b	67.46a	74.64b	81.32a
Média	10.23	10.28	96.91	96.92	1076	1076	59.16	60.00	74.64	75.52
CV (Tx A)	6.44%		0.55%		0.17%		1.41%		0.19%	
F K-S	0.994	0.054	2.266	0.123	4.43	0.418	1.775	0.09	2.424	0.055
p (Tx A)	0.0005		0.0001		0.0000		0.000		0.0000	
Age	Shape Index (%)		Shell index (g/100 cm ²)		EWMT (mm)		EWOMT (mm)		EMT (mm)	
	T1	T2	T1	T2	T1	T2	T1	T2	T1	T2
27	75.71	74.96	8.42	8.65	0.318	0.318	0.240a	0.243a	0.077	0.077
31	76.46	76.09	8.73	8.68	0.294	0.308	0.237a	0.232a	0.063	0.068
36	77.28	77.55	8.73	8.84	0.278	0.283	0.228a	0.224a	0.051	0.064
40	77.03	76.56	8.34	8.55	0.278	0.276	0.226a	0.224a	0.051	0.055
46	77.21	77.14	8.87	8.92	0.287	0.298	0.251a	0.251a	0.048	0.047
48	77.50	76.87	8.5	8.93	0.280	0.289	0.241b	0.248a	0.039	0.041
53	76.02	76.11	8.93	9.03	0.285	0.292	0.252b	0.266a	0.035	0.032
58	77.18	76.65	9.1	9.41	0.308	0.317	0.248b	0.272a	0.037	0.043
63	75.01	75.76	8.99	8.43	0.302	0.283	0.257a	0.243b	0.041	0.037
Média	76.61a	76.43a	8.74a	8.82a	0.292a	0.296a	0.242	0.245	0.050a	0.052a
CV (Tx A)	4.20%		6.45%		8.02%		3.64%		28.75%	
F K-S	1.508	0.055	1.032	0.053	1.254	0.034	2.738	0.038	1.471	0.04
p (Tx A)	0.9947		0.0603		0.2453		0.000		0.4343	

¹Averages followed by different letters in the line into each variable, differ between them by Tukey test at 0.05 of significance; ²C.V.: Experimental variation coefficient; ³F: K-S: Levene and Kolmogorov-Smirnov test statistic; values in black indicate homogeneous variance and residues with regular distribution, respectively, both at 0.01 of significance; ³P (T x A): probability of the statistic of Snedecor test.



In general, the eggs had the same specific gravity, independent of egg weight, except those from 48 and 63-week-old breeders, where increasing and constant weight eggs resulted in higher specific gravity, respectively (1.076 and 1.077). The mineral matter percentage in the eggshells did not vary with the eggs weight, except with 31, 36 and 46-week-old breeders, whose eggs of greater weight presented lower mineral matter percentage. Mineral matter and specific gravity were stable, with little variation among the repetitions, according to age and egg weight, confirmed by the low coefficients of sensu Pimentel-Gomes (2000).

Egg volume and eggshell surface were directly related to egg weight, while egg shape index ($p=0.9947$) and eggshell ($p=0.0603$) were not dependent, being the averages variation close to 76.5% and 8.78g, 100 cm², respectively.

The eggshell with membrane and membrane thickness did not follow the eggs weight among the treatments. For shell without membrane thickness there was a difference among treatments over 48 weeks of age, and from 48 to 58 weeks of age, the lower weight (T1) eggs had lower thickness, and at 63 weeks of age, there was an inversion of this behavior.

Characteristics of eggshell quality and breeders' age are in Figure 1. The percentages of eggshell weight of both treatments over time were dispersed, not fitting to the models tested (Figure 1A), while the percentage of mineral matter of the shells remained constant (between 95.87 and 97.5%) with the advancement of time (Figure 1B).

For specific gravity, an extreme point in constant and increasing eggs from 27-week of age birds was seen and, therefore, the decreasing exponential model was adjusted. From 31 weeks of age on (Figure 1C), the curves were similar, remaining practically constant up to 63 weeks ($R^2=94.04\%$ and 95.06% respectively).

The breeder age advancement had low impact on the egg shape index (Figure 1D), not adjusting to the regression models tested by the unpredictability of response. However, for both treatments, low rates can be observed in early ages followed by an increase up to around 46 weeks and decreasing values from that age, however this variable presents low variation throughout the bird life (75 to 77.5%).

Egg volume (Figure 1E) and shell surface (Figure 1F) at T2 presented weekly linear increases of 0.3751 cm³ and 0.3219 cm², respectively, with the age advancement (R^2 above 95%). These variables show values almost constant for T1, confirming the direct relation of these characteristics with the egg weight

without, however, being influenced by the age, and did not fit to any regression curve. The amount of shell deposited per area (Figure 1G) did not fit to the regression models, and when analyzing the dispersion, it is noted that most of the data present a linear increasing trend with age in both treatments suggesting an increase of shell deposition due to the age increase. Scattered spots were observed at 40, 48 and 63 weeks of age.

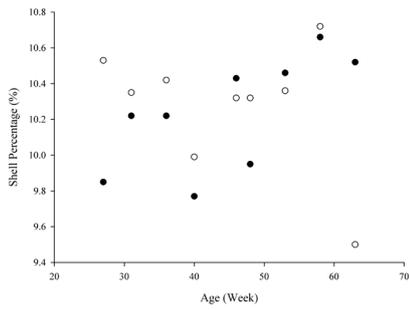
The eggshell with membrane thickness (Figure 1H) showed an adjustment to the quadratic model for eggs with constant weight ($R^2=77.7\%$), minimum point close to 44 weeks of age, reaching a minimum thickness of 0.280 mm. The eggshell thickness for increasing weight eggs, although not adjusting to any regression model ($R^2\geq 70\%$), the data tended to quadratic, with scattered points at 48 and 63 weeks of age. The eggshell without membrane thickness (Figure 1I) decreased until the 40th week old and from the 46th week on, both treatments increased. The eggshell membrane thickness of constant (T1) and increasing (T2; Figure 1J) weight eggs decreases 0.0010 and 0.0012mm with increasing age, $R^2=79.5\%$ and $R^2=87\%$, respectively.

DISCUSSION

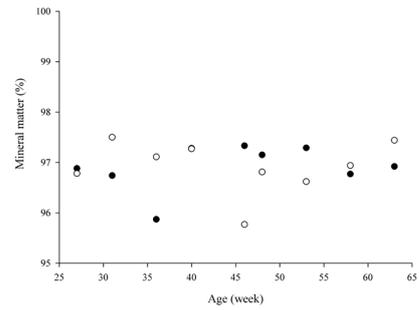
The egg volume and the eggshell surface area had the same behavior of egg weight for both treatments, that is, constant for treatment 1 and increasing for treatment 2. For Tumova & Ledvinka (2009), the eggshell quality decrease, with the advancing age, is related to the egg size and eggshell surface area increase. However, in this study, the same behavior among treatments was observed for eggshell quality.

The percentage deposition of mineral matter in the eggshell throughout the breeder life was practically constant for both treatments, indicating that the contents available to the eggshell are the same throughout the breeder life regardless of the egg weight. Although the eggshell index and percentage had shown an increasing linear trend with increasing age, the mineral matter percentage remained constant. This way, the transfer of minerals from the bird to the egg throughout the life does not compete for impairment in the eggshell quality.

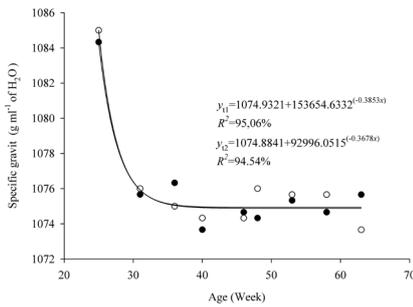
The specific gravity behavior related to egg weight demonstrates that this variable is constant, which corroborates with Frank *et al.* (1964) who found a low correlation ($R=0.14$) among these variables. The specific gravity along with the advancing age showed a drop from 27 to 31 weeks of age, possibly due to



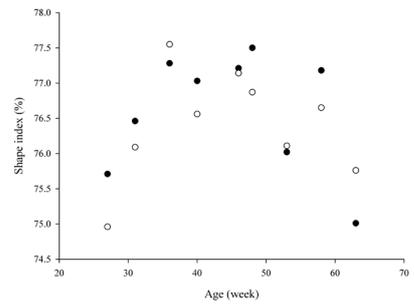
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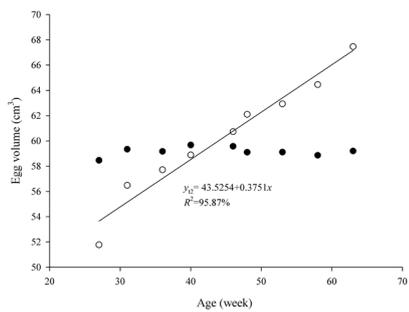
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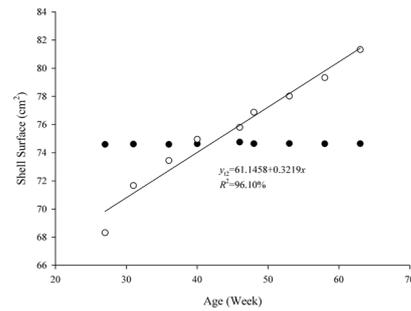
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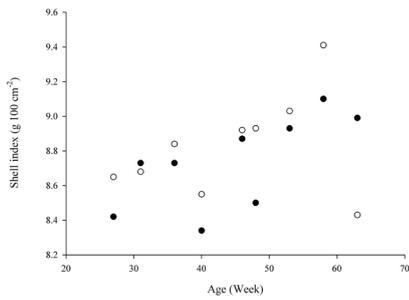
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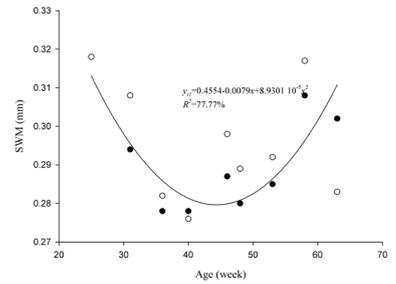
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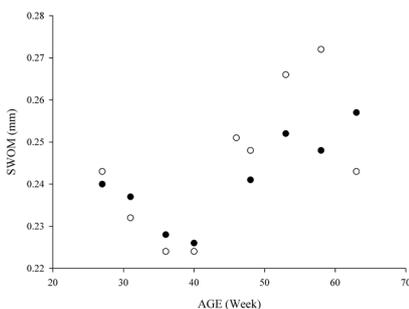
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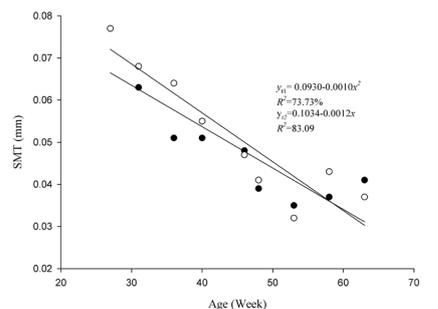
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H



I



J

Figure 1 – Description and quality of constant (T1●) and increasing (T2○) eggshell mass from Cobb 500 broiler breeders. Eggshell percentage (A), Mineral matter (B), Specific gravity (C), Shape index (D), Egg volume (E), Eggshell area (F) and eggshell index (G), EWM – Eggshell thickness with membrane (H), EWOM – Eggshell thickness without membrane (I) e EMT – Eggshell membrane thickness (J).



the beginning of the birds' egg production, but from 31 to 63 weeks of age, it remained constant, which could be attributed to the genetic evolution of the studied lineage. Similar behavior, with reduction at the beginning of the posture and maintenance in the next phase was observed in the same lineage (Kontecka *et al.*, 2012). For Tumova & Gous (2012) the interaction of four factors (bird type, age, oviposition and temperature) affects more intensely the specific gravity, than a single factor. As the eggs of this experiment came from the same lineage, same laying date, temperature and collection time, we can infer that the factors that influenced the specific gravity were minimized, confirming to be a constant variable independent of the breeder age gain or of the egg increasing weight.

The eggshell thickness is not influenced by the egg weight, which corroborates with Wolanski's *et al.* (2007) findings, of low correlation ($R = 0.02$). Regarding age, the thickness decreases up to 48 weeks of age and presents increasing values from this age on. This result can be related to the breeders feeding management with feed amount weekly reduced, with the same nutritional levels from the beginning of the laying until 46 weeks old, concomitant with laying high index and persistence. However, following the company's nutritional program, after 48 weeks of age another nutritional formulation is provided, with an increase in the daily calcium supply, coinciding with the period that the breeder is reducing the laying index (Table 1).

The eggshell membrane thickness decreases with the breeder increasing age. The membrane thickness decrease would be related to the physical barrier quality decrease (Mine, 2003), in addition, this reduction can affect the eggshell structure (Barbosa *et al.*, 2012).

In general, hatching eggs are oval shaped according to the shape index (SI of 74.96 to 77.55%) which is independent of the weight, probably the genetic selection of the lineage sought to maximize the use of eggs for incubation. According to Sarica & Erensayin (2009), round and long eggs do not fit well on the cartons, being more prone to cracking and breaking during transport and handling. The egg shape index throughout the breeder life, although presenting close values, showed a quadratic tendency, that is, until half of the breeder life, the eggs tend to be subtly rounded and from that phase on, elongated. Similar results to the one reported by Tumova & Gous (2012) in a research with the same lineage (Cobb), with eggs from 36 and 64-week-old breeders, found a change in the shape index, with the birds' age increase, being older breeders the ones that produce longer eggs.

CONCLUSION

Cobb 500 broiler breeders increasing age affects the eggshell quality, independent of the egg's weight, maintaining the mineral matter concentration constant. Thus, the eggshell quality decrease with the advancing age cannot be attributed solely to the egg weight, volume and shell surface increase, but also to geriatric factors.

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