NATIONAL AGRICULTURAL AND FOOD CENTRE
RESEARCH INSTITUTE FOR ANIMAL PRODUCTION NITRA

Slovak J. Anim. Sci., 50, 2017 (1): 1–6

ISSN 1337-9984

EVALUATION OF SOME EGGSHELL PARAMETERS DURING THE EMBRYOGENESIS IN TURKEYS

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ABSTRACT

The aim of the experiment was to investigate the traits of eggshells of incubated turkey eggs, produced by turkeys at 34 and 46 weeks of age and to evaluate their effect on the development of embryos. This study was performed in the turkey farm of the Poultry and Rabbit Selection, Population Genetics and Technology unit at the Agricultural Institute – Stara Zagora in 2016. The eggs from turkeys of the North-Caucasian Bronze (NCB) breed were investigated. One hundred and twenty eggs were randomly collected from 34 and 46-week-old turkeys. The eggs were examined by the 9th day of incubation and before the transfer to the hatcher. The numbers of dead embryos and their eggshell parameters were registered. The shells of eggs of viable hatchlings were also analysed. Egg weight, shell weight, shell thickness (at sharp end, blunt end and equator), number of pores, egg surface area and shell density of eggs with embryos dead and hatched from 34 and 46-week-old turkeys were investigated. At 34 and 46 weeks of age, the weight of eggshells of eggs with early dead and late dead embryos was significantly higher (8.35 g at $p < 0.001$ and 8.14 g at $p < 0.01$) compared to the eggshell weight of hatched eggs - 7.27 g. There were no differences in the thickness of shells of dead and hatched eggs, laid by 34- and 46-week-old turkeys. The total number of pores on the shell surface of early dead eggs was lower when compared to the parameter of late dead eggs and hatched eggs from turkeys at 34 and 46 weeks of age. Eggs with dead embryos had thicker shells than hatched eggs in turkeys at the two studied ages (34 and 46 weeks of age). This requires further studies on the influence of the quality of the egg shell on the development of embryos.

Key words: turkey egg; shell; porosity; thickness; incubation

INTRODUCTION

The thickness and porosity of eggshells are among the most important factors influencing the hatchability of eggs (Tsarenko and Kurova, 1989; Narushin's and Romanov, 2002). For successful embryonic development and hatching, an optimum number of pores distributed properly on the eggshell surface, of specific length with regard to the adequate water and gas exchange, is necessary (Burton and Tullett, 1983; Christensen, 1983; Burton and Tullett, 1985).

The eggshell thickness and the amount of pores differ among the bird species. In general, the optimum number of pores per cm^2 of turkey eggs is 51-59 $n.cm^{-2}$, and shell thickness is about 0.37 mm. Pore diameter varies from 0.01 to 0.04 mm, with smaller base and larger surface opening (Dyadichkina *et al.*, 2014)

For chicken eggs, the total number of pores is about 8000 vs. 5000 for turkey eggs (Burtov *et al.*,1990). Water and gas exchange is directly related to egg porosity, so the latter is closely related to the intensity of the embryonic development. The hatchability of eggs with both low and high porosity is low, and viability of hatchlings is decreased (Chistyakova, 1988).

In a research on the quality of eggs from meat type chickens, Gafarova and Nuriev (2014) established that eggshells of studied eggs was 0.35 mm thick, the number of pores on the sharp end of eggs was from 6300 to 7800, whereas on the blunt end - from 11200 to 12500.

Christensen (1983) demonstrated that the age of layer had a substantial effect on eggshell porosity. Pores were more numerous in eggs laid during the first production week as compared to those laid during the 10th week and by the end of the production cycle.

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Received: November 22, 2016
Accepted: February 3, 2017

The author found a relationship between the spatial distribution of pores on eggshell surface and the good embryonic development of chicks and hatching, but not with the amount of pores. Epimahova (2010) also found a correlation between the number of pores on eggshells with the age of layers. In the beginning of lay, the number of pores in turkey eggs was by 25 to 50 per 1 cm^2 lower than those during peak production.

Besides the number of pores, thickness and weight are other eggshell features. Peeblesand Brake (1985) reported that broiler chicken eggs were the thickest also with greatest embryonic death rates. Gerzilov (2011) detected differences in eggshell thickness in the studied chicken genotypes. Sharlanov *et al.* (1988) reported increased hatchability from 67 to 85 % parallel to increase in turkey eggshell thickness from 0.44 to 0.50 mm. On the contrary, Andrews (1972) observed higher hatchability of turkey poults from eggs with thinner shells. Numerous authors (Kostova, 1974; Shatokhina, 1975; Kurova, 1986) reported higher embryonic death rate in eggs with relatively thick and thin shells compared to embryonic death rates of eggs with medium thickness. However, Malik *et al.* (2015) did not observe any statistically significant effect of eggshell thickness on the hatchability traits of broiler chicken eggs.

Eggshell quality is influenced by the age of the laying hens (Zabudskii, 2016). In turkey eggs (Ghane *et al.*, 2015), quail eggs (Genchev, 2014) and chicken eggs (Petrov *et al.*, 2011), the eggshell percentages were reported to decrease as the age of layers increased. The thickness of eggshells together with the shell membrane, as found out by Hristakieva *et al.* (2009), was 0.44 mm in eggs laid by 32-week-old turkeys and 0.43 mm in eggs from 44-week-old turkeys. Mróz *et al.* (2014) also noticed lower eggshell thicknesses as turkeys became older. Sharipkulova *et al.* (2012) reported higher eggshell thickness and density (from 1.0700 ± 0.0009 to 1.078 ± 0.2200) in 80-week-old Lohmann White layers compared to that of layers at 26 weeks of age.

The purpose of the present experiment was to investigate the traits of eggshells of incubated turkey eggs, produced by turkeys at 34 and 46 weeks of age and to evaluate their effects on the development of embryos.

MATERIAL AND METHODS

The experiments were performed in the stud turkey farm of the Poultry and Rabbit Selection, Population Genetics and Technologies unitat the Agricultural Institute – Stara Zagora in 2016.

The eggs from turkeys of the North-Caucasian Bronze (NCB) breed were investigated. The birds were reared in the stud farm on deep permanent litter at a density of 3 $birds.m^{-2}$. They were fed standard ration for turkey layers containing metabolizable energy - 2987.17 kcal, crude protein 18.10 %, calcium 2.8 7 %, available phosphorus 0.49 %. Average daily feed intake was 300 g.

One hundred and twenty eggs were randomly collected from 34-week-old turkeys. Every egg was numbered and weighed before the incubation, which took place in Optima incubators. The eggs were examined by the 9^{th} day of incubation and before the transfer to the hatcher. The numbers of dead embryos and their eggshell parameters were registered.

The shells of eggs of viable hatchlings were also analyzed. Similarly, eggs from turkeys at 46 weeks of age were studied.

The measurement of egg and shell weights was done with a precision of 0.01 g on a balance. The shell thickness was determined with a micrometer. The number of pores was evaluated with methylene blue staining (0.5 g 89 % dye in 1 L of 70 % ethanol) pipetted on shell surface, left to impregnate the pores for better visibility and staining (Board and Halls, 1973). The number of pores was counted under a 2.5 × magnifying glass in four 0.25 cm^2 squares, in each studied zone (sharp end, blunt end, equator). The average density of pores per 1 cm^2 was determined as mean arithmetic of four measurements per zone (Peebles and Brake, 1985).

The total number of pores on eggshell surface was calculated by multiplication of the average number from the three studied zones (sharp end, equator and blunt end) to the eggshell surface area.

The egg surface area (cm^2) - SA was calculated by the formula (Carter, 1975):

$SA\ (cm^2) = 3.9782 \times EW^{0.7056}$,

where, EW - egg weight (g)

The shelldensity ($mg.cm^{-3}$) SD was calculated by the formula (Curtis *et al*., 1985):

SD ($g.cm^{-3}$) - Shell weight (g) / [(surface area, cm^2) × (shellthickness, cm)]

Data were analysed using descriptive Statistics, t-Test: two-sample as suming equal variances using Excel 2003-ANOVA (Zhelyazkov and Tsvetanova, 2002).

RESULTS AND DISCUSSION

The incubation traits of eggs from turkeys at studied ages (Fig. 1) demonstrated lower fertility of eggs laid by 34-week-old birds (by 7.5 % compared to 46-week-olds). During the embryogenesis of eggs of older turkeys, the percentages of early dead and late dead embryos were higher (7.2 and 9.90 %, respectively). The hatchability of set eggs and fertile eggs was greater in eggs from younger turkeys (81.67 % and 66.67 % vs. 85 % and

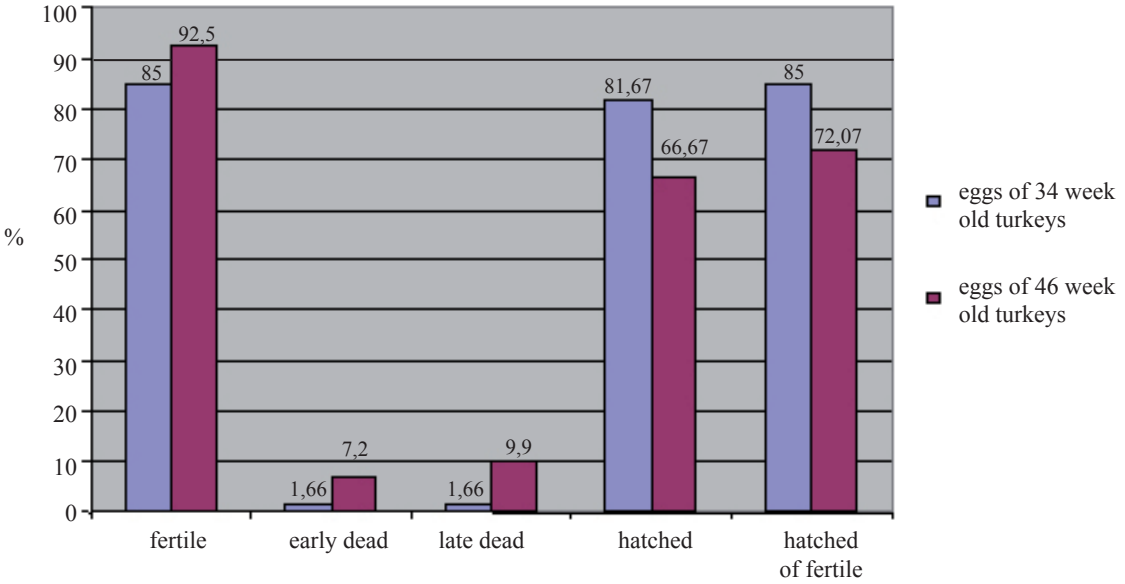

Fig. 1: Incubation traits of eggs from turkey hens at 34 and 46 weeks of age

72.07 %, resp. in eggs from 34- or 46-week-old hens).

Table 1 presents the values of studied shell parameters of incubated eggs from 34-week-old turkeys. The egg weights were the highest in eggs with embryos dead between incubation days 1 and 9 (88.23 g). At later incubation stages, there were eggs with lower weight observed (79.01 g). The eggs, from which viable poults were hatched, have average weight of 81.69 g. Eggshell weights in eggs with embryos dead during incubation were significantly higher ($p < 0.001$) than in eggs that hatched: 9.86 g in eggs with early dead embryos, 8.69 g in eggs with late dead embryos and 6.94 g in hatched eggs.

There were not significant differences in eggshell thickness between early dead and late dead eggs, as well as in hatched eggs, except for thickness measured in the equator of late dead eggs, which turned out to be significantly higher ($p < 0.01$): 0.40 mm compared to early dead and hatched eggs (0.36 and 0.36 mm,

Table 1: Eggshell parameters of incubated eggs from turkeys at 34 weeks of age (mean ± SD)

Parameters	Egg type		
	Early dead	Late dead	Hatched
Egg weight (g)	$88.23 \pm 1.400^{a***}$	79.01 ± 1.540^{b}	81.69 ± 0.390^{b}
Shell weight (g)	$9.86 \pm 0.510^{a***}$	$8.69 \pm 0.330^{a***}$	6.94 ± 0.050^{b}
Shell thickness (mm)			
Sharp end (SE)	0.36 ± 0.030	0.38 ± 0.020	0.39 ± 0.010
Equator (E)	0.36 ± 0.050^{a}	$0.40 \pm 0^{b**}$	0.36 ± 0.010^{a}
Blunt end (BE)	0.39 ± 0.050	0.37 ± 0.010	0.37 ± 0.010
Average shell thickness (mm)	0.37 ± 0.010	0.38 ± 0.003	0.37 ± 0.008
Number of pores SE ($n.cm^{-2}$)	23.50 ± 2.000	23.00 ± 7.000	31.15 ± 4.070
Number of pores E ($n.cm^{-2}$)	33.00 ± 5.000^{a}	42.50 ± 15.500^{ab}	$58.38 \pm 5.910^{b**}$
Number of pores BE ($n.cm^{-2}$)	30.50 ± 3.500^{a}	39.50 ± 2.500^{ab}	$53.61 \pm 2.450^{b*}$
Total number of pores (n)	2739.66 ± 389.540^{a}	3051.83 ± 647.750^{ab}	$4242.79 \pm 292.650^{b**}$
Egg surface area (cm^2) - SA	$93.80 \pm 0.560^{a***}$	86.82 ± 1.960^{bc}	89.15 ± 0.840^{c}
Shell density ($g.cm^{-3}$) - SD	$2.88 \pm 0.090^{a***}$	2.60 ± 0.360^{ab}	2.10 ± 0.020^{b}

Different letters (a, b, c) within a row indicate statistically significant differences: *$p < 0.05$; ** $p < 0.01$; ***$p < 0.001$

respectively). Higher embryonic death rates during the early embryogenesis were observed by Peebles and Brake (1985) in eggs with thick shells. The higher thickness of shells reduced its permeability; hen embryonic death could be anticipated due to the unfavourable effect of the two factors.

The numbers of pores in the three studied shell zones (sharp end, equator, blunt end) were significantly ($p < 0.05$) higher in hatched eggs compared to those in eggs with dead embryos. The average number of pores in hatched eggs was 31.15 n.cm^{-2} at the sharp end, 58.38 n.cm^{-2} at the equator and 53.61 n.cm^{-2} at the blunt end.

The same tendency was remained for the total amount of pores on egg surface; it was the highest in hatched eggs (4242.79), significantly ($p < 0.01$) lower in early dead eggs (2739.66) and late dead eggs (3051.83). Similar data were reported by Burtov *et al.* (1990).

The surface area of eggs was the highest in early dead eggs (93.80 cm^2, $p < 0.001$) compared to both late dead and hatched (86.82 cm^2 and 86.15 cm^2, respectively). This is attributed to the higher weight of such eggs, as surface areas is closely related to egg weight.

In the present study, there was positive correlation between eggshell density and eggshell weight, thickness and egg surface area. The highest SD values were observed in early dead eggs (2.88 g.cm^{-3}), which had also higher eggshell weight (9.86 g). Lower eggshell density was exhibited by eggs with late embryonic death (after the 10th day of incubation) and in normally hatched eggs: 2.60 g.cm^{-3} and 2.10 g.cm^{-3} respectively ($p < 0.001$).

The results of incubated eggs from turkeys at 46 weeks of age are presentedin Table 2. During that part of the production cycle, not significant differences were found in the weight of eggs between dead and viable embryos. The eggshell weight was significantly higher in early and late dead eggs ($p < 0.001$ and $p < 0.01$, respectively) compared to that of hatched eggs.

In this study, the eggshell thickness values were not substantially different in dead and viable eggs, while a number of other researchers (Kostova, 1974; Shatokhina, 1975; Kurova, 1986) demonstrated lower embryonic death rates in eggs with intermediate eggshell thickness compared to thicker or thinner eggshells.

Pores of shells were the most numerous in the equator region of hatched eggs (59.73; $p < 0.001$) compared to those in early and late dead eggs. The results of present study are not consistent with those of the study of Peebles and Brake (1985), as authors reported higher density of pores in the blunt end of hatched eggs.

In this study, the total number of pores on the surface of eggs with dead embryos in the early incubation period was the lowest (4133.43) followed by hatched eggs (4871.12) and eggs with embryos dead between the 9th and 25th days of incubation (5101.91). The eggs with extreme porosity, either very low or very high,were with poor hatchability, and hatchlings – with reduced viability (Chistyakova, 1988).

The surface area of hatched eggs was 90.87 cm^2, i.e. significantly ($p < 0.05$) lower than that of late dead embryos (91.87 cm^2) and slightly lower than that of early dead embryos (91.48 cm^2).

The shell density varied. In early dead eggs it was 2.31 mg.cm^{-3}, vs. 2.24 mg.cm^{-3} in late dead eggs and 2.10 mg.cm^{-3} in hatched eggs.

Table 2: Eggshell parameters of incubated eggs from turkeys at 46 weeks of age (mean ± SD)

Parameters	Egg type		
	Early dead	Late dead	Hatched
Egg weight (g)	85.11 ± 1.250	85.61 ± 1.770	84.30 ± 0.790
Shell weight (g)	8.35 ± 0.210^{a***}	8.14 ± 0.320^{a**}	7.27 ± 0.080^{b}
Shellt hickness (mm)			
Sharp end (SE)	0.40 ± 0.010	0.39 ± 0.010	0.39 ± 0.006
Equator (E)	0.39 ± 0.008	0.39 ± 0.012	0.38 ± 0.005
Blunt end (BE)	0.38 ± 0.010	0.39 ± 0.012	0.37 ± 0.005
Average shell thickness (mm)	0.39 ± 0.006	0.39 ± 0.009	0.38 ± 0.005
Number of pores SE (n.cm^{-2})	36.42 ± 3.400^{a***}	55.66 ± 4.580^{b}	40.80 ± 2.500^{a**}
Number of pores E (n.cm^{-2})	42.83 ± 4.840^{a}	51.83 ± 8.640^{ab}	59.73 ± 3.970^{b***}
Number of pores BE (n.cm^{-2})	55.17 ± 6.730	59 ± 5.390	56.53 ± 3.250
Total number of pores (n)	4133.43 ± 364.920	5101.91 ± 348.830	4871.12 ± 209.470
Egg surface area (cm^2) - SA	91.48 ± 0.940^{ab}	91.86 ± 0.310^{ab*}	90.87 ± 0.370^{a}
Shell density (g.cm^{-3}) - SD	2.31 ± 0.040	2.24 ± 0.090	2.10 ± 0.030

Different letters (a, b) within a row indicate statistically significant differences: *$p < 0.05$; ** $p < 0.01$; ***$p < 0.001$

Figure 2 depicts the total number of eggshell pores of early dead, late dead and hatched eggs from turkey hens at 34 and 46 weeks of age. The number of pores of shells of older turkey hens was higher both in dead and hatched eggs. This is in line with the data reported by Gupalo (2014), but disagrees with the result of Kontecka *et al.* (2012), who did not observe any significant differences in the porosity of shells of eggs laid by hens at a various age. Szczerbińska (1997) reported lower number of pores on shells of eggs from chickens in peak production.

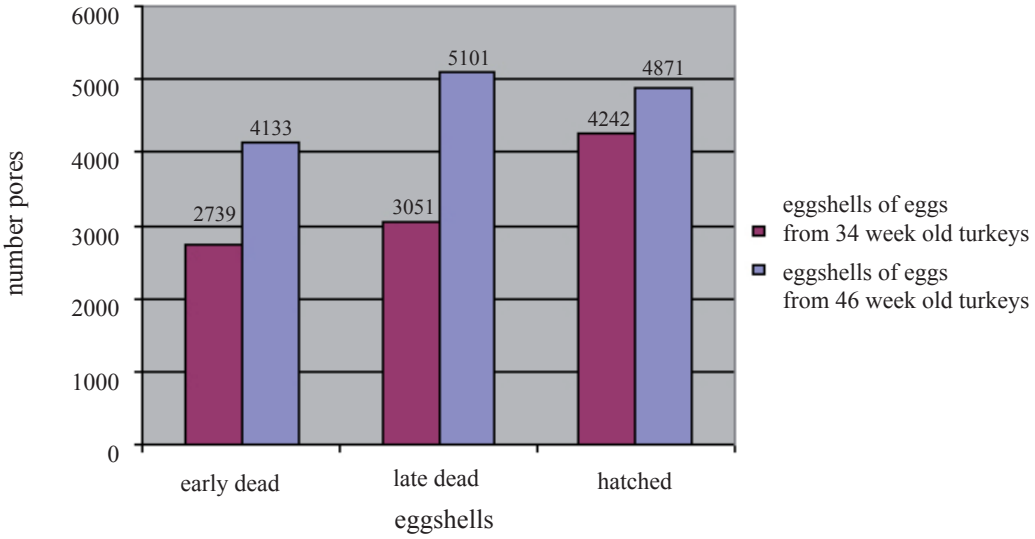

Fig. 2: Total number of eggshell pores of eggs from turkey hens at 34 and 46 weeks of age

CONCLUSION

At 34 and 46 weeks of age, the weight of eggshells of eggs with early dead and late dead embryos was statistically significantly higher ($p < 0.001$ and $p < 0.01$) compared to the eggshell weight of hatched eggs. There were no differences in the thickness of shells of dead and hatched eggs, laid by 34- and 46-week-old turkeys. The total number of pores on the shell surface of early dead eggs was lower when compared to the parameter of late dead eggs and hatched eggs from turkeys at 34 and 46 weeks of age. The eggs with dead embryos had thicker shells than hatched eggs in turkeys of the two studied ages (34 and 46 weeks of age). This requires further studies on the influence of the quality of the egg shell on the development of embryos.

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