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# Comparison of Lohmann White and Lohmann Brown Strains in Embryo Physiology

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Abstract: Chicken post-hatch performance is known to be related to embryonic developmental parameters. However, strain or genotype differences with regard to embryo physiological parameters have received little attention. A total of 1,200 hatching eggs produced by Lohmann Brown (LB) and Lohman White (LW) breeders of the same age were studied. Between 62 and 150 h of incubation, eggs Resonance Frequency (RF) was measured as indicator of early embryonic development. Also, albumen pH was measured between setting and d 8 of incubation. From d 10 to 18 of incubation, remaining albumen and embryos were weighed. During the last days of incubation, hatching occurrences were monitored after every four hours and hatched chicks were recorded. Results indicate that RF of LW eggs were lower than that of LB eggs (p<0.01) and starting time point of RF decrease occurred earlier in LB eggs than in LW eggs. Albumen pH of LB eggs was lower than that of LW eggs at day 8 of incubation. Remaining albumen weight at 14 and 16 d of incubation was lower in LB than in LW (p<0.05) while embryo weights increased more rapidly in LB strain than in LW strain. It is concluded that LB and LW embryos have different growth trajectories and should be incubated at different conditions.

**Key words:** Genetic line, embryo physiology, hatching events

## INTRODUCTION

During the past five decades, intensive selection in layers has focused on egg production rate and feed conversion in order to achieve increased productivity. But, all layer strains do not have similar physiology and/or development trajectories. It is well known that broiler post-hatch performance is related to embryonic developmental parameters (Tona et al., 2003). Similarly, the effects of incubation conditions, egg storage conditions and age of breeders on embryonic parameters are well known (O'Dea et al., 2004; Tona et al., 2004). However, layer strain or genetic line differences with regard to embryo physiological parameters have received little attention. Crittenden and Bohren (1961), Siegel et al. (1968) and Suarez et al. (1997) showed variations in incubation time with genotype. These variations in incubation time suggest that perhaps optimum incubation conditions need to be established for different genotypes or strains to obtain optimal hatching performance and high-quality chicks. Within the layer strains used for egg production, there are mostly two different types for brown or white eggshell. For instance, in Lohman and Hissex layers there are brown and white types. This difference in eggshell color suggests genotype difference and therefore may lead to differences in embryo growth trajectory as well as embryo physiology. Also, it can be

hypothesized that this genotype difference may influence incubation duration when brown and white type hatching eggs are incubated in similar conditions. To our knowledge, there is no information about comparison of embryonic parameters of layer strains or genetic lines differing in egg shell color.

The aim of this study was to compare (1) embryonic developmental parameters using acoustic resonance methods, (2) embryonic physiological parameters and (3) the hatching spread of eggs from Lohman Brown (LB) and Lohman White (LW) breeders of the same age.

## **MATERIALS AND METHODS**

Experimental design: A total of 1,200 hatching eggs, provided by Levrau Hatchery Company (Tielt, Belgium), were used for this study. Half (600) of the eggs were collected from a commercial flock of LB layer breeders and the other half (600) were from a commercial flock of LW layer breeders. The age of both layer breeders' strains was 45 weeks. The hatching eggs were stored for 7 days before setting for incubation. Prior to setting for incubation, eggs were numbered, weighed, assigned into 4 replications of 150 eggs each/strain. The eggs were incubated in Petersime® Vision incubator at 37.6°C, relative humidity of 50% and turning each hour through an angle of 90°. Prior to setting for incubation and also between 66 and 130 h of incubation, samples

of eggs were used to measure changes in resonant frequency as indicator of early embryonic development. During incubation, eggs were randomly set in order to avoid incubator gradients. From day 10 to 16 of incubation, albumen utilization by embryos was measured. At d 18 of incubation, eggs were candled and those with evidence of living embryos were transferred from turning trays to hatching baskets. During the last two days of incubation, hatching occurrence of individual egg was monitored after every two hours and hatched chicks were recorded.

Measurement of embryonic development: Embryo development was monitored by acoustic resonance analysis as described previously by Coucke et al. (1997). Briefly, the method involved the mechanical excitation of the egg by a mechanical impacter. The impactor hit the egg at its equator and the noise of the vibrating egg was recorded by a microphone positioned at the equator at an angle of 90° to the impacter. The recorded signal was then sent to a data acquisition card and transformed by fast Fourier transformation to obtain the resonant frequency for the first spherical mode of the vibrating egg. In this experiment, four instantaneous excitations with a phase shift of 90° were applied at the equator zone of the eggs. Samples of 150 eggs per strain were used to determine Resonant Frequency (RF). The measurements were done prior to setting for incubation and were repeated every 4 h between 66 and 130 h of incubation.

Albumen and embryo weighing: At d 10, 12, 14, 16 and 18 of incubation, samples of 45 eggs per strain were broken to determine weights of the growing embryos and the amount of remaining albumen. After breaking each egg, its embryo was removed carefully and separated from all attachments such as yolk sac and chorioallantoic membrane. The embryo was then wiped with absorbent paper before weighing. Also for each egg, the remaining thick albumen was separated from the embryonic fluid and weighed.

Hatching occurrence: Between 490 h and 518 h of incubation, the transferred eggs were checked every 4 h for the occurrence of hatching. At hatching stages, incubation duration was defined as the time between setting and the occurrence of this event for an egg. Spread of hatch was defined as the dispersion around the average incubation duration.

Statistical analysis: The data were processed with the statistical software package SYSTAT. The generalized linear regression model was used to analyze the effects of strain on albumen weights, albumen pH, resonant frequency and incubation duration. When the means of the general model were statistically different, they were compared using Tukey's test.

In a second analysis, hatchability was considered as binomial in distribution. A two-tailed test for comparison of variances was used to analyze the influence of strain on hatchability.

#### **RESULTS**

Effects of strains on resonant frequency: Figure 1 shows egg RF variations between 62 and 150 h of incubation according to strain. From 62 until 130 h of incubation, the RFs of LB eggs were lower than those of LW eggs (p<0.01). In both strains, RF increased slightly between the 62<sup>th</sup> h and 98<sup>th</sup> h of incubation and then decreased sharply. Although RF decrease followed a similar trend in both strains, the starting time point of decrease was earlier in the LB eggs (94<sup>th</sup> hr) compared to that of LW eggs (98 h) (p<0.05).

Strain effect on albumen utilization, albumen pH and embryo growth during incubation: Irrespective of strain, Fig. 2 indicates that albumen pH during early stage of incubation increased between egg setting time and day 2 of incubation and then decreased until day 8 of incubation. Up to day 6 of incubation, albumen pH was similar between strains. But, at day 8 of incubation, average albumen pH of LB eggs was significantly lower than that of LW eggs (p<0.01).

Figure 3 shows changes in albumen weights of the eggs of different strains at different stages of incubation. At day 10 and day 12 of incubation, albumen weights were not different between strains. Between day 12 and day 16 of incubation, there was a rapid decrease in albumen weight in both strains. However, at day 14 and day 16 of incubation, albumen weights were lower in LB eggs than in LW eggs (p<0.05). At day 18 of incubation, albumen weight was at nadir level in both strains.

Embryo weights according to incubation period and strain are shown in Fig. 4. Irrespective of strain, embryo weights increased from day 10 to 18 of incubation

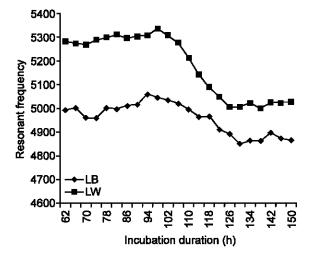


Fig. 1: Changes in Resonant Frequency (RF) according to incubation duration and strains

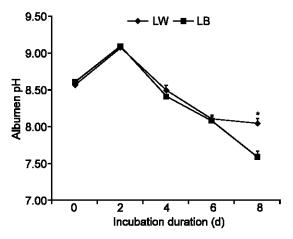


Fig. 2: Albumen pH according to incubation day and strains. At each incubation day, \* indicates difference between albumen weights.

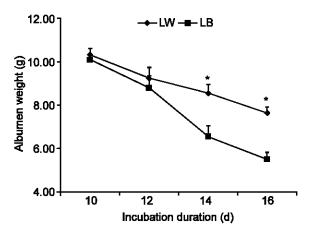


Fig. 3: Remaining albumen weights according to incubation day and strains. At each incubation day, \* indicates difference between albumen weights

(p<0.001). From day 12 of incubation onward, embryos from LW eggs had lower weights than those from LB eggs (p<0.01).

Effects of strains on hatching events: Figure 5 shows the distribution of hatching times according to strain. The hatching curve shows that the majority of LB eggs hatched between 490 and 506 h of incubation reaching a peak at 502h, whereas for eggs LW most hatching started to increase at 506h to reach a peak at 514h. The figure thus shows a delay in the hatching of LW eggs compared with LB eggs. At the end of hatch, average incubation duration of LB chicks was significantly lower than that of LW chicks of about 7h (p<0.01).

# DISCUSSION

Overall, a differential embryonic developmental pattern between both lines could be related to differences in

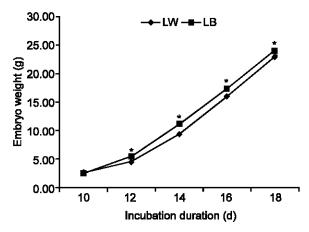


Fig. 4: Embryo growth in relation to post-hatch stage and according to strains. At each incubation day, \*indicates difference between albumen weights

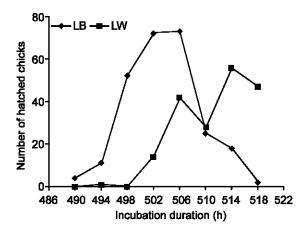


Fig. 5: Hatching curve in relation to the incubation duration and strain

hatching process. During the whole incubation period, LB had a faster development than LW. This faster embryonic growth in LB resulted in shorter incubation duration of about 7 h than that of LW strain.

Higher RF of LW eggs compared to LB eggs during incubation may be due to differences in eggshell characteristics. This observation is in line with the findings of De Ketelaere et al. (2002) who reported differences in eggshell characteristics between 6 different lines of chickens. Bamelis et al. (2002) had shown that changes in resonant frequency can be related with embryo fluid formation during early embryonic development and hence is a marker for a well defined developmental stage. The earlier general decrease in the RF of the egg of LB strain (at 94 h of incubation) compared to the LW eggs (at 98 h of incubation) indicates a quicker initiation of embryonic development, which might have been correlated with shorter incubation duration for this strain. Kemps et al. (2003) previously reported a positive correlation between the timing of RF decrease during early incubation and

hatching time. Consequently, at the same incubation time point, LB embryos and LW embryos may not have been at a similar physiological stage of development.

The same as RF, lower albumen pH of LB eggs at d 8 of incubation also indicates quicker initiation of LB embryo. Indeed, change in egg albumen pH during early embryonic stage is related to the rate of embryo initiation and growth. Because the yolk remains slightly acid, an almost 1000-fold hydrogen ion concentration gradient exists across the blastoderm in its position between albumen and yolk (Stern, 1991). Thus, changes in the viscosity or pH of the albumen may play a role in determining the viability of the embryo during early stages of development (Benton and Brake, 1996). Interestingly, between d 14 and d 16 of incubation, remaining albumen weight was lower in LB strain than in LW strain suggesting that during this stage LB embryo utilize faster albumen for growth. Indeed, during incubation, albumen proteins move into the amniotic fluid and are swallowed by the embryo. These proteins are then either digested in the gut of the embryo for growth or transferred into the yolk sac where they can be utilized after hatching (Deeming, 1989).

The faster growth of LB embryo compared with LW embryo observed up to d 16 of incubation lasted until end of incubation. As result, incubation duration was 7 h shorter for LB eggs than that of LW eggs. This delay in hatching of LW eggs can be related to a delay in the initiation of embryogenesis (Becker *et al.*, 1968; McLaury and Insko, 1968; Mather and Laughlin, 1976) and in a decrease in rate of embryo development as shown in RF and albumen data.

It can be concluded that LB and LW embryos have differential growth trajectories. These different patterns of growth are deduced from differences in physiological parameters. These findings suggest that differences in physiological parameters during embryonic development and also in physical parameters of the eggs may lead to the hypothesis that incubation conditions could be improved in a strain- dependent manner.

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