Effects of mixing eggs of different initial incubation time on the hatching pattern, chick embryonic development and post-hatch performance

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ABSTRACT

Background. The hatch window that varies from 24 to 48 h is known to influence post-hatch performance of chicks. A narrow hatch window is needed for commercial poultry industry to acquire a high level of uniformity of chick quality. Hatching synchronization observed in avian species presents possibilities in altering hatch window in artificial incubation.

Methods. Layer eggs of different initial incubation time were mixed on day 12 of incubation. During the hatching period, hatching time of individual eggs and hatch window were obtained by video cameras. Embryonic development and post-hatch performance up to day 7 were measured.

Results. The manipulation of mixing eggs of different initial incubation time shortened the hatch window of late incubated eggs in manipulated group by delaying the onset of hatching process, and improved the hatchability. Compared to control groups, chick embryos or chicks in egg redistribution group showed no significant difference in embryonic development and post-hatch performance up to day 7.

Discussion. We have demonstrated that eggs that incubated with advanced eggs performed a narrow spread of hatch with higher hatchability, normal embryonic development as well as unaffected chick quality. This specific manipulation is applicable in industrial poultry production to shorten hatch window and improve uniformity of chick quality.

INTRODUCTION

Hatching synchronization is of importance to precocial avian species that can be achieved by acceleration (*Holmberg 1991; Vince 1964*) or retardation (*Persson & Andersson 1999; Vince 1968*) of hatching. This adaptive advantage enables the offsprings to avoid being abandoned by motherhood and exposure to predators (*Davies & Cooke 1983*). In commercial poultry incubation, hatching synchrony essentially contributes to the uniformity of newly hatched chicks. In general, hatch window as evaluation of degree of hatching synchrony, which is defined as the time between early-hatching and late-hatching, varies from 24 to 48 hours (*Careghi et al. 2005; Decuypere et al. 2001*). Thus, early hatched chicks will be held in the incubators with deprivation of feed and water until entire batch of chicks hatch rather than removed immediately upon hatching. Variability of delayed time in feed access caused by the spread of hatch depressed uniformity of post-hatch performance of the chicks, including organ development, immune system activation, digestive enzyme stimulation and relative growth post hatch (*Tona et al. 2003; Willemsen et al. 2010a*).

In artificial incubation, the inherent characteristics of eggs (e.g. parental age, egg weight and egg storage time) and incubation conditions (temperature and CO_2 concentration during hatching phase) play a crucial role in embryonic development that results in the spread of hatch (*De Smit et al. 2006; Ipek & Sozcu 2017; Maatjens et al. 2014; Nangsuay et al. 2016; Tona et al. 2003; Tona et al. 2007; Willemsen et al. 2010b*). Moreover, intraclutch hatch synchronization was found in lesser snow goose (*Davies & Cooke 1983*), pheasants and mallard ducks (*Persson & Andersson 1999*), by shortening or prolonging the incubation period. In addition to this effect of sibling contact, the hatch process could also be affected by mixing eggs of different embryo developmental trajectory (*Tona et al. 2013*). However, no efficient manipulations during incubation have been performed to shorten hatch window in poultry production.

Therefore the aim of the present study was to achieve a narrowed hatch window through the manipulation of mixing eggs of different growth curves. In addition, potential effects on embryonic development and post hatch performance were studied. Hatching time of individual chicks, hatch window, hatchability, yolk residue and organ weights, body weight and leg bone development were compared between control and manipulated groups.

MATERIALS AND METHODS

All procedures in this study were approved by committee of the Care and Use of Animals of Zhejiang University, Hangzhou, China.

Experimental design

Hatching eggs (n=704; weight range from 53 to 57 g) were obtained from a Hyline breeder flock at 35 to 36 wk of age (Shenhai Breeding, Shenyang, China). The eggs were divided into early incubation group (EI) and late incubation group (LI), respectively. LI started incubation 12 hours later than EI so that the biological age (BA, calculated from the initial incubation time) of LI was 12 h older than that of LI. On BA 12 d of LI (BA 12.5 d of EI), 160 eggs randomly chosen from both EI and LI were distributed into the third incubator that defined as manipulated incubation group (MI). And the remaining eggs in EI and LI were regarded as control groups. The eggs in MI were distributed randomly. On BA 18 of LI, MI group were separated into EMI group (early incubated eggs in MI) and LMI group (late incubated eggs in MI) to start hatching phase.

Incubation

Eggs were incubated and hatched in lab-scale incubators (NK-hatching, Dezhou Nongke Incubation Equipment Co. Ltd., Shandong, China) measuring 1100×1000×900 mm with a capacity of 352 eggs. The incubators were calibrated by a standard thermometer and hygrometer before egg incubation. The incubation maintained at a temperature of 37.8 ± 0.1 °C and a relative humidity around 60%. The turning time interval during incubation was two hours until day 18. Eggs were candled (Cool-Lite tester, GQF) on day 18 and those with a living embryo were transferred to hatching baskets. Fisheye cameras (DS-2CD3942F-I, HIKVISION) adopted upon the hatching baskets were used to monitor the hatch process. All incubations stopped at BA 504 h of LI and chicks were removed from the incubators.

Post-hatch housing and management

A total of 128 newly hatched chicks (32 per incubation group) were sampled and transferred to four pens of 1 m^2 covered with sawdust. Artificial lightning was set for 23 h/d from day 0 to 7 (40 lux at chick eyes level). Temperature was set to 34°C, decreased by 0.5°C per day during 7 days. Feed and water were provided *ad libitum*.

Data collection

On BA 18 d and 20 d of LI, six eggs or chicks that hatched at peak hatching period (30% to 70% hatch) were randomly sampled from each groups for measurements of chick embryonic development. After eggs broken open, embryos or chicks were sacrificed by decapitation to obtain yolk weight and yolk free body weight (YFBW). Weights of heart, liver and stomach (gizzard and proventriculus) of all sampled embryos sacrificed on BA 18 d and 20 d of LI were determined.

The hatching time of individual eggs were determined using video recordings. And the hatching time is presented as biological age. From the first hatchling, quantity of chicks was obtained every an hour. The chicks were removed from incubators every twelve hours to allow the camera to get a clear field of view. Hatch window was calculated by subtracting hatching time of the last chick from that of the first chick. The peak hatching period was defined as 30% to 70% hatch of the batch.

At BA 504h of LI (516 h of EI), 32 chicks per group which hatched in peak hatching period were sampled and weighed. Metatarsus length (ML) was measured for assessment of leg bone development. After seven days' growing, all chickens received same measurements to evaluate post-hatch development.

Statistical analysis

A one-way ANOVA model (SPSS 19.0) was used to analyze the effects of egg redistribution on the embryonic development of chicks (Yolk free body weight, yolk weight, heart weight, liver weight and stomach weight) and post hatch performance (chick weight and tibia length). The level of significance was set at $P < 0.05$. The Fisher's LSD method was performed to test for overall differences among treatment groups. All data are shown as average ± S. E. M.

RESULTS

Hatch performance

The distribution of hatching time was obtained by video recordings of four treatment groups. The EI group was found to give the first hatchling as expected, and the hatch window was 38 hours (Figure 1. a). However, hatch process of EMI group started 5 hours later than EI, while it finished at the same time as EI group (Figure 1. b). The start-up time of egg incubation in LI and LMI groups were 12 hours later than those of EI and EMI. As a result, first chicks of LI and LMI groups emerged from eggs 2 and 8 hours later than EI group, respectively. The hatch process of LI group lasted 30 hours (Figure 1. c), 8 hours shorter compared to EI group. Moreover, LMI group had a shortened hatch window of 21 hours with highest hatchability (95.8%), even though it started at 468 h (Figure 1. d) which was 6 hours later than LI. According to 30% and 70% hatch time in Figure 2, the peak hatching period of manipulated incubation groups (EMI: 472.3 -478.8 h; LMI: 475.0 – 480.4 h) was delayed 1.9 to 2.7 hours compared to the control groups (EI: 470.4 -477.0; LI: 472.1 – 478.7). Furthermore, the duration of peak hatching period in EMI was shortest (5.4 hours) which was consistent with the narrow hatch window.

Embryonic development from day 18 until hatch

Embryonic development of four groups on BA 18 d of LI was shown in Table 1. Yolk free body weight was higher than in early incubation groups (EI and EMI) than late incubation groups (LI and LMI), but yolk weight of early incubated eggs (EI and EMI) was found significantly lower than late incubated eggs (LI and LMI). In addition, organ size (heart weight and liver weight) was larger in EI and EMI, mainly caused by higher YFBW. Whereas, no significance of stomach weight was found.

The four incubation groups hatched in peak hatching period had similar YFBW (Table 2). Due to the earlier peak hatching period of chicks in four groups, yolk absorption of EI was faster and those chicks had higher liver and stomach weight. And LMI chicks that had short holding time in incubator hatched with significantly higher yolk weight, lower liver and stomach weight. However, heart development of all hatched chicks was similar in all groups.

Overall, there were no significant differences between EI and EMI or LI and LMI in YFBW, yolk absorption and organ size. No effects of egg redistribution were observed for embryonic development both on BA 18 d and 20 d of LI.

Post-hatch performance until day 7

The evaluation of post-hatch performance until day 7 was presented in Table 3. At peak hatching time of LMI (480 h), body weight of chicks in early incubation groups (EI and EMI) was lower due to weight loss in holding period in hatchers, while EI and EMI chicks had higher ML. Whereas, no significant difference was found between EI and EMI, as well as LI and LMI. Similar results occurred after seven days' growth. Although both body weight and ML of early incubation groups (EI and EMI) were slightly higher than those of late incubation groups (LI and LMI), post hatch growth and leg bone development was not altered by the manipulation of egg redistribution.

DISCUSSION

The aim of this study was to investigate the effects of egg redistribution during incubation on hatching time and post-hatch development. The results demonstrate that mixing eggs of different developmental stages during incubation influenced hatching process, including delayed hatching time and shortened hatch window. They also suggest that embryonic development and post-hatch performance were not altered by egg redistribution on BA 12 d of LI.

Hatching time is known to be influenced by factors such as parental age, egg storage time and conditions, and incubation conditions (*Careghi et al. 2005; Decuypere & Bruggeman 2007; Tona et al. 2003*). And hatching time distribution results in different chick qualities and physiological characteristics in one batch of hatched chicks (*Careghi et al. 2005; Wang et al. 2014*). To eliminate these factors, eggs were obtained from a single breeder flock, laid on the same day, stored with very short time (no more than 2 days), and incubated in incubators with temperature and relative humidity calibration. Thus, the manipulation of egg redistribution on day 12 might be the only factor that affect the hatching time in this study.

The present study confirmed that mixing eggs of different growth curves shortened the hatch window of redistribution group, which is consistent with hatching synchronization found in pheasants (*Persson & Andersson 1999*). The onset of hatching process in redistributed eggs was retarded 5 to 6 hours, indicating that the narrow hatch window was related to delay of the first hatch in manipulated group. This might be explained by some kinds of egg communication between early incubated eggs and late incubated eggs. Chick embryos begin to develop a functionary auditory system as early as day 10 of incubation (*Alladi et al. 2002*). Specific interaction among redistributed eggs may take place after mixing eggs, by means of embryo sound communication. Perception of vocalizations by embryos may lead to physiological or behavioral changes. This confirms the finding of Tong (2015) that delayed internal piping time when embryos were exposed to embryo sound stimulation. However, an increased mortality was observed in duck and chicken eggs that were incubated under artificial sound stimulation (*Tong et al. 2015a; Veterany et al. 1999*). Compared to the artificial sound stimulation, embryo vocalization may lay less stress on other hatching eggs that exerts no negative impact on hatchability. Another hypothesis is that environmental CO_2 altered the hatch process and results in narrow spread of hatch. Previous researches reported that high levels of CO_2 during early stages of incubation stimulated early hatching and shortened hatch window (*De Smit et al. 2006; Tona et al. 2007*). Although the onset of hatching process of mixed eggs was delayed compared to the control groups, this did not extend the spread of hatch. The early incubated embryos may penetrate the membrane and eggshell, generate more CO_2 during hatching period, leading to increasing CO_2 concentration that stimulated the hatching process of late incubated eggs. Furthermore, the increasing concentration potentially contributes to hatchability of LMI (95.8%) - higher than the other groups - suggests that more chick embryos succeeded in breaking out of eggshell rather than died in this tough process. Considering this delayed onset of hatching process, the narrow spread of hatch and the increased hatchability, our future work will focus on identifying as to what degree and via which mechanisms, redistributing eggs of different growth curves affects hatching pattern and hatchability.

The advanced embryonic development of early incubated eggs was observed in both control (EI) and manipulated group (EMI), mainly caused by the initial incubation time difference of 12 hours. However, mixing eggs of different growth curves did not alter the embryonic growth and yolk absorption before hatch. Chick embryos of both early incubated and late incubated eggs were able to maintain normal organ development and nutrient metabolism until hatch. Although earlier hatched chicks (EI and EMI) underwent a longer holding period in incubators, the decreased yolk weight and increased organ weight indicated an advanced maturation of organs post hatch, as supposed by previous studies (*Pinchasov & Noy 1993; Tong et al. 2015b; Van de Ven et al. 2011*). Long time no access to feed and water (EI and EMI, 36 h; LI and LMI, 24 h) resulted in a higher weight loss in early incubation groups (EI and EMI), but enhanced the leg bone development. The consistency of body weight and leg bone development on day 7 was observed as expect. Nevertheless, the narrow hatch window of manipulated groups did not influence chick growth performance up to day 7, indicating that egg distribution only stimulates the hatching behavior. However, there is no evidence that response to eggs or egg communication by egg distribution was related to this shortened hatch window. As above, there was no negative effect of mixing eggs of different growth curves on embryonic growth, utilization of nutrients and post-hatch performance.

CONCLUSION

The specific manipulation of mixing eggs of different initial incubation time influenced hatching pattern, including onset of hatching process and hatch window. The egg redistribution during incubation did not affect normal embryonic development, utilization of nutrients and post-hatch performance. All of these results will be applicable in industrial hatchery to shorten hatch window and improve uniformity of chicks.

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