Available online at www.sciencedirect.com
ScienceDirect
Comparative Biochemistry and Physiology, Part A 147 (2007) 529–535
CBP
www.elsevier.com/locate/cbpa

Endothermic heart rate response in broiler and White Leghorn chicks (*Gallus gallus domesticus*) during the first two days of post-hatch life

H. Yoneta [a], E.M. Dzialowski [b], W.W. Burggren [b], H. Tazawa [a,*]

[a] *Department of Electrical and Electronic Engineering, Muroran Institute of Technology, Muroran 050-8585, Japan*
[b] *Department of Biological Sciences, P.O. Box 305220, University of North Texas, Denton, Texas 76203, USA*

Received 25 October 2006; received in revised form 1 February 2007; accepted 1 February 2007
Available online 9 February 2007

Abstract

The embryonic modal value of heart rate (MHR) differs between broiler and White Leghorn chickens, but the initial development of cholinergic chronotropic control of embryonic heart rate (HR) does not. Thus, we hypothesized that hatchling MHR should also differ between broiler and White Leghorn strains, while the development of a physiological regulation, such as the endothermic HR response, should not be different between hatchlings of the two strains. To test this, we measured the response of HR and cloaca temperature (Tb) to alteration of ambient temperature (Ta); *i.e.*, 35 °C–25 °C–35 °C, in four groups of hatchlings on Days 0 and 1 post-hatch. Fertile eggs of both strains with similar mass were incubated simultaneously in the same incubator. Eggs of broiler chickens hatched ~7 h earlier than White Leghorn chicken eggs. Chick mass at hatching was identical in both strains, but diverged during 2 days after hatching. Tb measured at the initial Ta of 35 °C was identical in both strains. MHR at the same Ta was ~30 bpm lower in broiler chicks than in White Leghorn chicks, but the difference was reversed to that observed in the embryos. The endothermic HR response was advanced by ~1 day in broiler chicks compared with White Leghorn chicks. As a result, eggs of similar mass in both strains produced chicks with similar mass and Tb at hatching, but during 2 days of post-hatch life their masses diverged and regulation of the endothermic HR response developed earlier in broiler than in White Leghorn hatchlings. This physiological heterochrony between strains is most likely due to genetic selection for fast growth in broiler chickens.

Keywords: Broiler chicken; Cloaca temperature; Endothermic heart rate response; Hatchling; Instantaneous heart rate; Mode heart rate; White Leghorn chicken

1. Introduction

The development of endothermy in avian embryos involves maturation of the metabolic machinery and cardiovascular system to support an increase in oxygen uptake (Tazawa and Hou, 1997; Tazawa and Whittow, 2000; Black and Burggren, 2004; Dzialowski et al., 2007; Seebacher et al., 2006). Given the cardio-respiratory physiology of adult strains of chickens varies considerably, it is likely that physiological heterochrony exists in the developmental timing of regulatory pathways involved in respiratory and cardiovascular control between broiler and White Leghorn chickens — two strains that have been selectively bred for different juvenile and adult phenotypes. Broiler chickens have been selected for increased rapid growth, while White Leghorn chickens were selected for a slower growing egg laying adult phenotype (Ohta et al., 2004; Zhao et al., 2004).

Development of cholinergic chronotropic control of embryonic heart rate (HR) had been reported to differ among a number of studies (Höchel et al., 1998; Crossley and Altimiras, 2000; Crossley et al., 2003b; Aubert et al., 2004; Chiba et al., 2004). Differences in the strains of chicken used in the different experiments were considered to be a potential reason for the inconsistency between the studies (Crossley et al., 2003a,b). Thus, Yoneta et al. (2006b) conducted an experiment to compare development of cholinergic chronotropic control of embryonic HR between two strains; broiler and White Leghorn chickens. Measurement of instantaneous heart rate (IHR) from embryos of both strains with similar mean fresh egg mass

* Corresponding author. Fax: +81 143 46 5501.
E-mail address: tazawa@mmm.muroran-it.ac.jp (H. Tazawa).

1095-6433/$ - see front matter © 2007 Elsevier Inc. All rights reserved.
doi:10.1016/j.cbpa.2007.02.001

showed that the initial development of cholinergic chronotropic control of embryonic HR was similar between broiler and White Leghorn chickens, but the modal value of embryonic IHR; *i.e.*, mode heart rate (MHR), determined over a 1-h period was significantly higher in broiler than White Leghorn chickens.

These physiological differences lead to a number of important developmental questions. First, given that embryonic MHR differs, is the MHR of broiler hatchlings also higher than that of White Leghorn hatchlings? Second, is the development of physiological function, such as endothermy, identical between newly hatched chicks of the two strains or do they exhibit physiological heterochrony (Spicer, 2006)? For newly hatched broiler chicks, we recently investigated development of endothermic HR response and found that HR response to 10 °C alteration of ambient temperature (Ta) switched from a temperature dependent change (*i.e.*, ectothermic thermo-conformity response) to an inverse temperature dependent change (*i.e.*, endothermic response) during the first two days post-hatch; *i.e.*, the day of hatching (Day 0) and Day 1 (Khandoker et al., 2004; Yoneta et al., 2006a). Therefore, it is hypothesized that the endothermic HR response in White Leghorn chickens should also develop during the first two days post-hatch; *i.e.*, Day 0 and Day 1, but the hatchling MHR of White Leghorn chickens should be lower than that of broiler chickens as observed in embryos. In order to examine the hypothesis, an experiment was designed to determine the time of hatching, changes in body mass after hatching, and responses of HR and cloaca temperature (Tb) to changes in Ta (ΔTa) on Days 0 and 1 post-hatch in broiler and White Leghorn chickens.

2. Materials and methods

Fertile eggs of broiler and White Leghorn chickens (*Gallus gallus*) were obtained from the same hatchery and Hokkaido Animal Research Center, respectively, as the previous experiment (Yoneta et al., 2006b). Eggs were weighed and numbered on the eggshell for identification. The mean egg mass of both strains was not significantly different. Eggs of both strains were simultaneously incubated at 38 °C and 55% relative humidity in a forced draft incubator as previously described (Yoneta et al., 2006b). A CCD camera was set in the incubator and hatching time of individual chicks was determined within 0.5 h by capturing video images of the hatching process. The mass of chicks was measured with a balance to 0.1 g when they hatched. Chicks were numbered on a tag attached to their legs and transferred to a brooding chamber maintained at 35 °C with food and water *ad libitum*. The body mass was measured again prior to experimentation and the body mass ratio between experimental mass and hatching mass was calculated as a measure of growth.

Table 1
Fresh egg mass, chick mass at hatching, ratio of chick mass and egg mass, and hatch time in broiler and White Leghorn chickens

	N	Egg mass (g)	Chick mass (g)	Chick/egg	Hatch time (h)
Boiler	122	67.4±0.4	49.2±0.3	0.730±0.003	494±0.7
White Leghorn	102	67.5±0.4	49.3±0.4	0.732±0.003	501±0.8
P		0.881	0.665	0.557	<0.0001

Mean±SE.

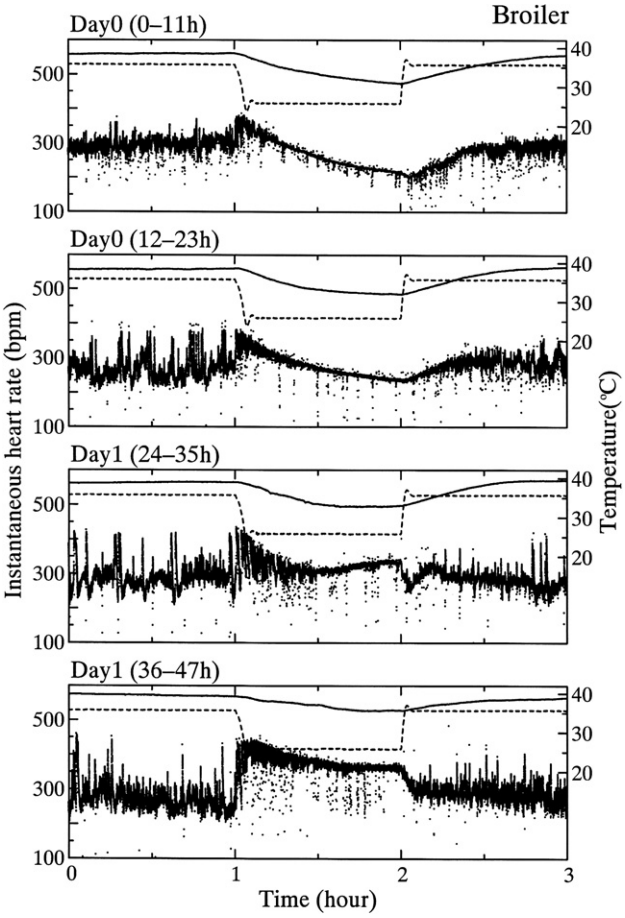

Fig. 1. Three-hour recordings of cloaca temperature (solid curve) and instantaneous heart rate (points) in hatchlings of broiler chickens exposed to ambient temperature of 35 °C, 25 °C and 35 °C for 1-h period each (dotted curve). From top downwards, hatchlings belonged to group 0–11 h on Day 0, group 12–23 h on Day 0, group 24–35 h on Day 1 and group 36–47 h on Day 1, respectively.

Two broiler and two White Leghorn hatchlings were simultaneously measured for HR responses to ΔTa with an experimental exposure sequence of 35 °C, 25 °C and 35 °C for 1 h each as previously described (Khandoker et al., 2004; Yoneta et al., 2006a). The measurement took at least 4 h, including an initial 1-h period for hatchlings to acclimate to a measurement chamber and temperature. Because the measurement was time-consuming and sometimes in vain due to unsuccessful recordings, incubation of eggs and experiments were made four times until an adequate number of data was obtained. The total number of chicks whose timing of the hatching process was identified was 122 and 102 individuals for broiler and White Leghorn chickens, respectively. Among them, 77 and 78 chicks were measured for responses of HR to ΔTa. Cloaca temperature (Tb) was simultaneously measured with a thin thermo-couple at a depth of about 1 cm as described previously (Tazawa et al., 2004). The HR and Tb were presented as the mean value taken over last

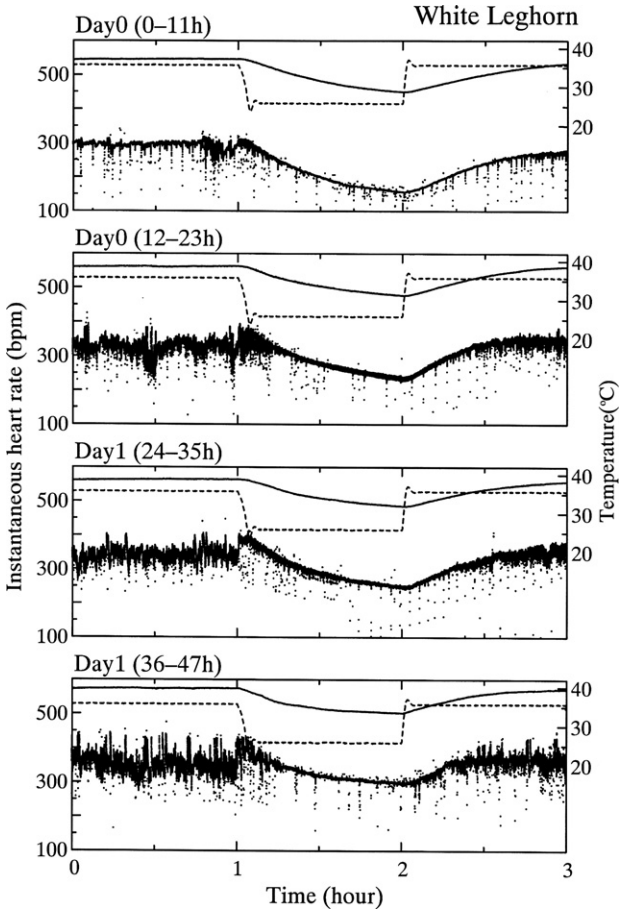

Fig. 2. Three-hour recordings of cloaca temperature (solid curve) and instantaneous heart rate (points) in hatchlings of White Leghorn chickens exposed to ambient temperature of 35 °C, 25 °C and 35 °C for 1-h period each (dotted curve). Groups of four panels are the same as in Fig. 1.

10-min recording for each individual exposure. Chicks were divided into four groups according to their age at the time of measurement; *i.e.*, those measured within the first 12 h post-hatch on Day 0 were divided into group 0–11 h on Day 0, and similarly group 12–23 h on Day 0, group 24–35 h on Day 1 and group 36–47 h on Day 1.

Student's unpaired *t*-test was used to examine for significant differences in mean values between 122 broiler and 102 White Leghorn chicks. Two-way factorial ANOVA with repeated measures with Tukey *post hoc* test examined the differences in MHR, Tb and body mass ratio between broiler and White Leghorn chicks and between the four developmental groups. The significance level was $P<0.05$. The mean values are shown with standard error (SE).

3. Results

Fresh egg mass, chick mass, ratio of chick mass to egg mass, and hatching time in both strains are summarized in Table 1. None of the mass parameters were significantly different between the two strains, but eggs of broiler and White Leghorn chickens hatched at 494 h and 501 h on average, respectively, and the difference was significant ($P<0.0001$).

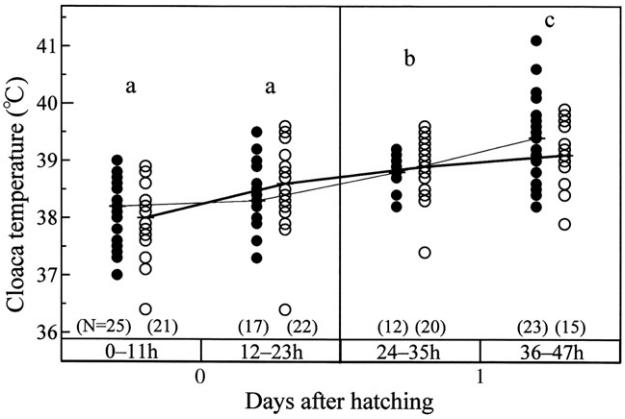

Fig. 3. Cloaca temperature (Tb) of individual hatchlings of broiler (closed circles) and White Leghorn (open circles) chickens, determined in 35 °C environment for four developmental groups. Numerical figure in the parentheses indicates the number of hatchlings examined. Thin and thick solid lines connect the mean value of Tb. A significant difference in group means between the four developmental stages is indicated by different letters.

Representative responses of Tb and HR in broiler hatchlings that belonged to the four developmental groups are shown in Fig. 1. A chick in group 0–11 h transiently increased HR upon exposure to Ta of 25 °C, but decreased HR in parallel with the decrease in Tb during the remaining cooling period. Similarly, an advanced chick on Day 0 (group 12–23 h) responded to cooling with a transient increase upon exposure and subsequent decrease during cooling, but the decrease was mitigated. On Day 1, chicks increased HR baseline upon cooling and maintained an elevated HR during the remaining exposure time. Fig. 2 shows responses of Tb and HR in White Leghorn hatchlings. In general, a transient increase in HR upon cooling was not predominant and HR baseline tended to decrease in parallel with Tb even in Day 1 embryos (groups 24–35 h and 36–47 h).

Tb of individual hatchlings of broiler and White Leghorn chickens, measured in the initial 35 °C environment in four developmental groups is shown in Fig. 3. Tb increased significantly on Day 1 in comparison with Day 0 ($P<0.01$).

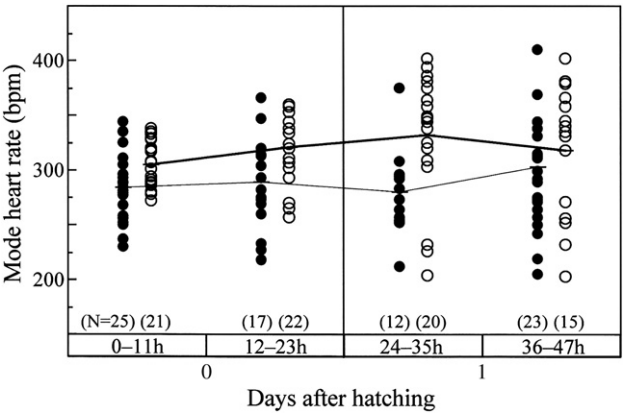

Fig. 4. Mode heart rate during 1-h period in individual hatchlings of broiler (closed circles) and White Leghorn chickens (open circles), determined in 35 °C environment for four developmental groups. Others are the same as in Fig. 3.

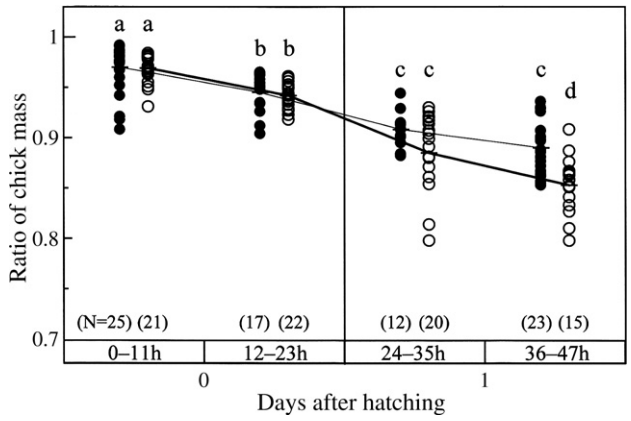

Fig. 5. Ratio of chick mass at each developmental stage to that at hatching in broiler (closed circles) and White Leghorn (open circles) chickens, determined for four developmental groups. Others are the same as in Fig. 3.

However, the difference in Tb between both strains was not significant ($P=0.975$). Fig. 4 presents MHR of individual chicks, measured at 35 °C in four developmental groups of broiler and White Leghorn chickens. Contrary to embryonic HR, hatchling HR of White Leghorn chickens was significantly higher than that of broiler chickens ($P<0.0001$). The difference in HR between the four developmental groups of both strains

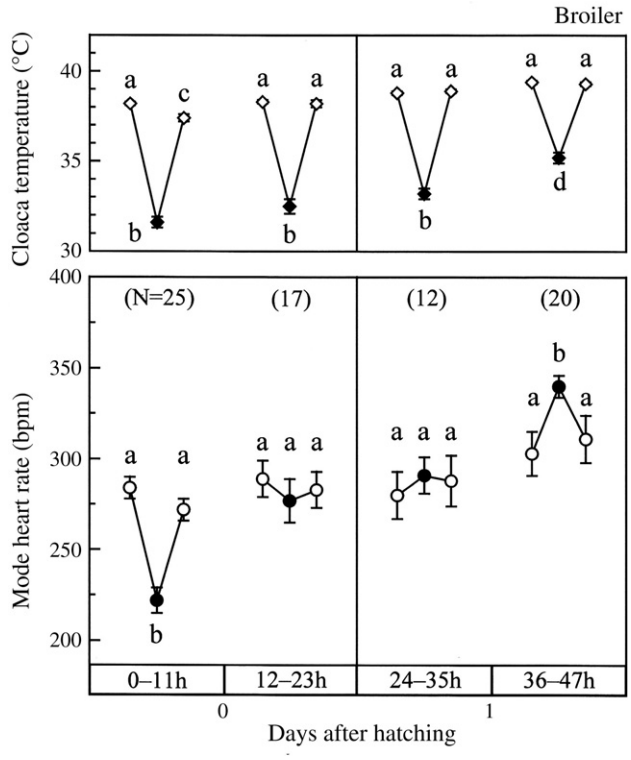

Fig. 6. Responses of cloaca temperature (top panel, diamonds) and mode heart rate (bottom panel, circles) to altered ambient temperature in hatchlings of broiler chickens at four developmental stages on Days 0 and 1 post-hatch. Numerical figure in the parentheses indicates the number of hatchlings examined. The mean values with SE are shown and solid lines connect the mean values. The significant difference between group means is indicated by different letters.

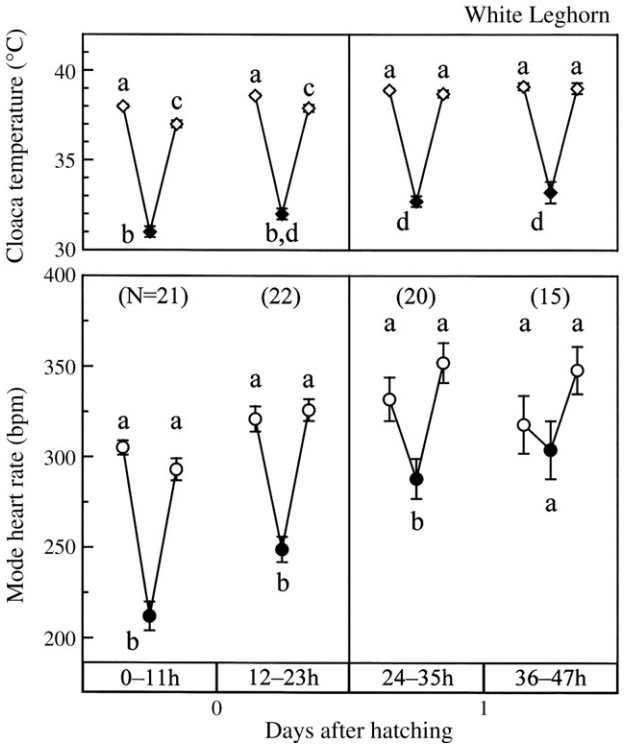

Fig. 7. Responses of cloaca temperature (top panel, diamonds) and mode heart rate (bottom panel, circles) to altered ambient temperature in hatchlings of White Leghorn chickens at four developmental stages on Days 0 and 1 post-hatch. Others are the same as in Fig. 6.

was not significant ($P=0.450$). Fig. 5 shows ratio of body mass at each developmental stage to that at hatching in individual chicks measured for their HR responses. The body mass ratio was significantly different between broiler and White Leghorn hatchlings ($P<0.0001$) and decreased significantly with progress of developmental stage.

Responses of Tb and MHR to Ta of 35 °C–25 °C–35 °C on Days 0 and 1 in hatchlings of broiler chickens are presented in Fig. 6. Tb at the initial Ta of 35 °C and that at the final Ta of 35 °C were significantly different on early Day 0 (group 0–11 h) ($P<0.01$), but did not differ in the other advanced chicks. On early Day 0, HR during cooling was significantly lower than that at 35 °C ($P<0.01$) and the HR response conformed to ΔTa; *i.e.*, thermo-conformity response. On late Day 0 (group 12–23 h) and early Day 1 (group 24–35 h), HR remained unchanged during cooling and warming exposures, which did not conform to ΔTa; *i.e.*, referred to as insufficient endothermic response. On late Day 1 (group 36–47 h), HR increased significantly during cooling ($P<0.05$) and decreased during warming ($P<0.05$) and an endothermic inverse temperature dependent change was observed. HR response to ΔTa switched from the thermo-conformity change on early Day 0 to an apparent endothermic pattern on late Day 1 in broiler chickens.

Responses of Tb and MHR to ΔTa, determined for hatchlings of White Leghorn chickens are shown in Fig. 7. Tb at the initial Ta of 35 °C and that at the final Ta of 35 °C were significantly different in both groups on Day 0 (groups 0–11 h and 12–23 h) ($P<0.01$). HR decreased significantly during

cooling in both groups on Day 0 and also the early group on Day 1 (group 24–35 h), thus showing a thermo-conformity response. On late Day 1 (group 36–47 h), the decrease in HR during cooling and the increase during warming were not statistically significant, resulting in an insufficient endothermic response at this stage in White Leghorn chickens.

4. Discussion

4.1. Initial development of endothermic heart rate response

The HR of developing avian embryos and hatchlings responds to altered Ta in accordance with the extent of development of thermoregulatory competence (Tazawa and Nakagawa, 1985; Tazawa et al., 1992, 2001, 2002; Ono et al., 1994; Tamura et al., 2003; Khandoker et al., 2004; Fukuoka et al., 2006; Yoneta et al., 2006a). The HR response to a cooling and warming exposure or a warming and cooling bout was previously determined to elucidate the development of thermoregulatory competence in chickens and emus (Tazawa et al., 2001; Tamura et al., 2003; Khandoker et al., 2004; Fukuoka et al., 2006; Yoneta et al., 2006a). In chickens, exposure to a cooling and warming bout revealed that externally pipped (EP) embryos showed a decrease in HR baseline during cooling, but advanced EP embryo that stayed inside an eggshell on Days 21 and 22 of incubation responded to cooling with an endothermic HR change (Tazawa et al., 2001). After hatching, the HR response to warming and cooling exposure in broiler chicks switched from thermo-conformity on early Day 0 to an endothermic inverse temperature dependent change on late Day 1 (Khandoker et al., 2004). Reversing the sequence of exposure; that is, cooling followed by warming, did not change the result for broiler chicks that responded with thermo-conformity HR change on early Day 0 maturing to an endothermic inverse temperature dependent response on late Day 1 (Yoneta et al., 2006a). From Day 2 onward, broiler hatchlings in both experiments exhibited complete endothermic HR responses (Khandoker et al., 2004; Yoneta et al., 2006a).

The present experiment was designed to confirm the broiler HR responses on Days 0 and 1 and compare this with White Leghorn hatchlings. In newly hatched broiler chicks, HR responses changed from thermo-conformity on early Day 0 to an endothermic inverse temperature dependent response on late Day 1. This involved passing through a stage of insufficient endothermic response, which confirmed the previous experiments with broiler chickens (Fig. 6; Khandoker et al., 2004; Yoneta et al., 2006a). In White Leghorn chickens, the HR response exhibited an ectothermic thermo-conformity on early Day 1 (group 24–35 h) and began to exhibit an insufficient endothermic response a half day later (group 36–47 h) (Fig. 7). Initially, we assumed that the initial development of endothermic HR response would not differ between broiler and White Leghorn hatchlings and would mature within two days post-hatch. However, developmental timing of the endothermic HR response differed between broiler and White Leghorn chickens and it appears to be advanced by one day in broiler chickens. These findings suggest that the mechanisms regulating the endothermic HR response exhibit physiological heterochrony between broiler and White Leghorn strains.

The HR of the developing embryo is controlled by both cholinergic and adrenergic inputs (Crossley and Altimiras, 2000; Aubert et al., 2004; Chiba et al., 2004; Yoneta et al., 2006b). The development of cholinergic chronotropic control of embryonic IHR in broiler and White Leghorn chickens was simultaneously investigated by determining IHR for 1-h period in embryos of both strains on Days 10, 11, 12, 13 and 14 of incubation (Yoneta et al., 2006b). The investigation revealed no significant difference in the early development of cholinergic HR control between the two stains. Similar findings were reported for broiler and bantam embryos (Crossley et al., 2003b).

The increase in HR during cooling observed in hatchlings exhibiting the endothermic response may be partially mediated by adrenergic pathways (Tazawa, 2005). Embryos of both White Leghorn chicken and emu exhibit a strong sympathetic tone during the later stages of development (Crossley and Altimiras, 2000; Crossley et al., 2003a). In White Leghorn embryos, this tone was mediated by circulating levels of catecholamines and not sympathetic efferents. It is possible that during the first days of post-hatch life, the sympathetic efferents mature and contribute to the endothermic HR response. The development of the adrenergic pathways after hatching may occur at different rates in the two strains, producing the physiological heterochrony in HR regulation. However, it remains to be determined if these differences are in fact due to differential maturation of the autonomic nervous system or humoral derived catecholamines.

While the primary control of embryonic IHR does not appear to differ between these two strains, embryonic MHR on Days 10–14 of incubation was significantly higher in broiler than in White Leghorn chickens. There was a switch sometime during development such that hatchling MHR was lower in broiler than White Leghorn chickens (Fig. 4). MHR of the broiler hatchlings measured in this study agrees with previous measurements of broiler hatchling HR, but is significantly lower than MHR of the present White Leghorn chickens (Khandoker et al., 2004; Yoneta et al., 2006a). This could be due to differences in either intrinsic HR or autonomic control of HR.

Another potential influence on MHR might be differences in Tb. The embryonic Tb of broiler chickens was reported to be higher than that of White Leghorn chickens due to high heat production in broiler chickens (Janke et al., 2004). Although high embryonic Tb of broiler chickens might be responsible for a high embryonic HR, hatchling Tb was identical between both strains and should not be responsible for the difference in hatchling HR. The reversal of HR when comparing embryos and hatchlings of both strains suggests that the developmental patterns (*i.e.*, daily changes) of HR during incubation and after hatching are different between both strains.

4.2. Mass change

In commercial breeding, broiler chickens were genetically selected to grow more rapidly than White Leghorn chickens during both incubation and post-hatch life (Ohta et al., 2004; Zhao

et al., 2004). The fast growth of broiler chicken embryos results in heavy body mass and may be associated with accelerated development of physiological regulation compared with White Leghorn chicken embryos after passing through identical initial development *in ovo*. Because the heavy broiler embryos are also produced by large eggs laid by large broiler hens, egg size may be a factor causing different development of embryonic physiological function. Thus, the present experiment was designed to ensure chicks hatched from eggs with the same initial mass. Although eggs of both strains were incubated simultaneously in the same incubator, eggs of broiler chickens hatched 7 h earlier than White Leghorn chickens (Table 1). This difference in hatching time was much shorter than the 1-day difference reported in a previous study using different egg mass of broiler (67 g) and White Leghorn (61 g) chickens (Janke et al., 2004), but the difference of 7 h between both strains was significant. The chick mass at hatching was the same between broiler and White Leghorn chickens and accordingly the chick mass to egg mass ratio was the same in both strains (Table 1). Even with the shorter incubation duration in broiler eggs compared with White Leghorn eggs, they both produced hatchlings with the same mass, suggesting that genetic selection for embryos to grow fast seems to work in the present broiler chickens.

The chick Tb increased significantly on Day 1, and the increase in Tb during four developmental stages was similar in broiler and White Leghorn chickens (Fig. 3). As a result, hatchlings of broiler and White Leghorn chickens that hatched from eggs with identical mass had similar mass and Tb at hatching. The body mass ratio in broiler and White Leghorn hatchlings on early Day 0 (group 0–11 h) was also identical; *i.e.*, 0.97. Because this ratio indicates body mass of a hatchling at the time of the experiment relative to initial mass at hatching, hatchlings in group 0–11 h lost 3% of mass. Although hatchlings could have free access to food and water, they lost about 6% of mass on late Day 0 (group 12–23 h) in both strains. Mass loss on early Day 1 (group 24–35 h) was 9% and 11 % in broiler and White Leghorn chickens, respectively, but the difference was not significant. However, on late Day 1 (group 36–47 h), broiler and White Leghorn chickens lost 11% and 15% of initial mass, respectively, and the difference was significant. Hatchlings of broiler chickens maintained their mass on Day 1, but those of White Leghorn chickens lost mass during Day 1. Previous studies showed that mass loss of broiler chickens during Days 0 and 1 was recovered subsequently (Khandoker et al., 2004; Tazawa et al., 2004). Broiler chickens seemed to take advantage of the yolk and food uptake more efficiently than White Leghorn chickens soon after hatching. This difference may reflect genetic selection for broiler chickens to grow fast compared with White Leghorn chickens, which may also provide broiler chickens with advanced development of endothermic HR response.

4.3. Conclusion

Differences between chicken strains have been observed in other physiological systems at the embryonic and hatchling stages of development. Strain differences in pulmonary artery relaxation (Martinez-Lemus et al., 1999, 2003), embryonic oxygen uptake (Kuenzel and Kuenzel, 1977; Sato et al., 2006), catecholamine metabolism (Saito et al., 2004), and growth (Ohta et al., 2004; Zhao et al., 2004; Sato et al., 2006) have all been observed. Broiler chickens have been selected for fast growth by rapidly producing muscle mass resulting in a muscular adult, while White Leghorn chickens have been selected for slower growth and egg laying ability. The artificial selection of differing physiological and morphological phenotypes of the growing juvenile and adult most likely produced the strain differences observed in the development of physiological regulatory mechanisms. While selection has been made on the adult phenotype, the differences begin to appear during early ontogeny.

Here we found that the development of hatchling endothermic HR response was advanced in broiler chickens compared with White Leghorn chickens. The mean fresh egg mass of both strains was not different. The mean chick mass at hatching was identical and accordingly the ratio of chick mass to egg mass was identical between broiler and White Leghorn chickens; *i.e.*, 0.73. In addition, Tb of hatchlings brooded at 35 °C at four developmental stages on Days 0 and 1 was identical between both strains. However, the eggs of broiler chickens hatched on average 7 h earlier than White Leghorn chickens, indicating statistically faster embryonic growth of broiler chickens compared with White Leghorn chickens even if egg mass was the same. In addition, even if chick mass at hatching was the same between both strains, chick mass on late Day 1 was already heavier in broiler chickens than in White Leghorn chickens, indicating fast hatchling growth of broiler chickens. The genetic selection for broiler chickens to grow fast compared with White Leghorn chickens was apparent even for chicks that hatched from the eggs with the same mass and the genetic selection seems to provide for earlier thermoregulatory competence in broiler chickens, reflected in the advanced development of an endothermic HR response in hatchlings of broiler chickens. These two strains exhibit physiological heterochrony in their development of endothermy which is most likely due to the artificial selection for rapid growth in the broiler stain.

Acknowledgments

This study was supported in part by National Science Foundation Grant No.IOB0417205 awarded to EMD and No. IBN0614815 to WWB and by a Grant-in-Aid for Scientific Research of the Japan Society for Promotion of Science (No.15560352) to HT. We are grateful to Ms. K. Kunishige, Hokkaido Animal Research Center, Shintoku, Hokkaido, for providing us with fertile eggs of White Leghorn chickens.

References

Aubert, A.E., Beckers, F., Ramaekers, D., Verheyden, B., Leribaux, C., Aerts, J.-M., Berckmans, D., 2004. Heart rate and heart rate variability in chicken embryos at the end of incubation. Exp. Physiol. 89, 199–208.

Black, J.L., Burggren, W.W., 2004. Acclimation to hypothermic incubation in developing chicken embryos (*Gallus gallus*): I. Developmental effects and chronic and acute metabolic adjustments. J. Exp. Biol. 207, 1543–1552.

Chiba, Y., Fukuoka, S., Niiya, A., Akiyama, R., Tazawa, H., 2004. Development of cholinergic chronotropic control in chick (*Gallus gallus domesticus*) embryos. Comp. Biochem. Physiol. A 137, 65–73.

Crossley, D., Altimiras, J., 2000. Ontogeny of cholinergic and adrenergic cardiovascular regulation in the domestic chicken (*Gallus gallus*). Am. J. Physiol., Regul. Integr. Comp. Physiol. 279, R1091–R1098.

Crossley, D.A., Bagatto, B.P., Dzialowski, E.M., Burggren, W.W., 2003a. Maturation Cardiovascular control mechanisms in the embryonic emu (*Dromaius novaehollandiae*). J. Exp. Biol. 206, 2703–2710.

Crossley, D.A., Hicks, J.W., Thornburg, K., 2003b. A comparison of cardiovascular regulatory mechanism present during fowl ontogeny; white leghorn *vs*. bantam chickens. Abstr. SICB, p. 147.

Dzialowski, E.M., Burggren, W.W., Komoro, T., Tazawa, H., 2007. Development of endothermic metabolic response in embryos and hatchlings of the emu (*Dromaius novaehollandiae*). Respir. Physiol. Neurobiol. 155, 286–292.

Fukuoka, S., Khandoker, A.H., Dzialowski, E.M., Burggren, W.W., Tazawa, H., 2006. Development of endothermic heart rate response in emu (*Dromaius novaehollandiae*) embryos. In: Yahav, S., Tzschentke, B. (Eds.), New Insights into Fundamental Physiology and Peri-natal Adaptation of Domestic Fowl. Nottingham Univ. Press, Nottingham, pp. 29–42.

Höchel, J., Akiyama, R., Masuko, T., Pearson, J.T., Nichelmann, M., Tazawa, H., 1998. Development of heart rate irregularities in chick embryos. Am. J. Physiol., Heart Circ. Physiol. 275, H527–H533.

Janke, O., Tzschentke, B., Boerjan, M., 2004. Comparative investigations of heat production and body temperature in embryos of modern chicken breeds. Avian Poult. Biol. Rev. 15, 191–196.

Khandoker, A.H., Fukazawa, K., Dzialowski, E.M., Burggren, W.W., Tazawa, H., 2004. Maturation of the homeothermic response of heart rate to altered ambient temperature in developing chick hatchlings (*Gallus gallus domesticus*). Am. J. Physiol., Regul. Integr. Comp. Physiol. 286, R129–R137.

Kuenzel, W.J., Kuenzel, N.T., 1977. Basal metabolic rate in growing chicks *Gallus domesticus*. Poult. Sci. 56, 619–627.

Martinez-Lemus, L.A., Hester, R.K., Becker, E.J., Jeffrey, J.S., Odom, T.W., 1999. Pulmonary artery endothemium-dependent vasodilation is impaired in a chicken model of pulmonary hypertension. Am. J. Physiol., Regul. Integr. Comp. Physiol. 277, R190–R197.

Martinez-Lemus, L.A., Hester, R.K., Becker, E.J., Ramirez, G.A., Odom, T.W., 2003. Pulmonary artery vasoactivity in broiler and Leghorn chickens: an age profile. Poult. Sci. 82, 1957–1964.

Ohta, Y., Yoshida, T., Tsushima, N., 2004. Comparison between broilers and layers for growth and protein use by embryos. Poult. Sci. 83, 783–787.

Ono, H., Hou, P.-C.L., Tazawa, H., 1994. Responses of developing chicken embryos to acute changes in ambient temperature: noninvasive study of heart rate. Isr. J. Zool. 40, 467–479.

Saito, S., Takagi, T., Koutoku, T., Saito, E.-S., Hirakawa, H., Tomonaga, S., Tachibana, T., Denbow, D.M., Furuse, M., 2004. Differences in catecholamine metabolism and behaviour in neonatal broiler and layer chicks. Br. Poult. Sci. 45, 158–162.

Sato, M., Tachibana, T., Furuse, M., 2006. Heat production and lipid metabolism in broiler and layer chickens during embryonic development. Comp. Biochem. Physiol. A 143, 382–388.

Seebacher, F., Schwartz, T.S., Thompson, M.B., 2006. Transition from ectothermy to endothermy: the development of metabolic capacity in a bird (*Gallus gallus*). Proc. Biol. Sci. 273, 565–570.

Spicer, J.L., 2006. A physiological approach to heterochrony. In: Warburton, S.J., Burggren, W.W., Pelster, B., Reiber, C.L., Spicer, J. (Eds.), Comparative Developmental Physiology: Contributions, Tools and Trends. Oxford University Press, Oxford, pp. 191–202.

Tamura, A., Akiyama, R., Chiba, Y., Moriya, K., Dzialowski, E.M., Burggren, W.W., Tazawa, H., 2003. Heart rate responses to cooling in emu hatchlings. Comp. Biochem. Physiol. A 134, 829–838.

Tazawa, H., 2005. Cardiac rhythms in avian embryos and hatchlings. Avian Poult. Biol. Rev. 16, 123–150.

Tazawa, H., Hou, P.-C.L., 1997. Avian cardiovascular development. In: Burggren, W.W., Keller, B. (Eds.), Development of Cardiovascular Systems: Molecules to Organisms. Cambridge Univ. Press, New York, pp. 193–210.

Tazawa, H., Nakagawa, S., 1985. Response of egg temperature, heart rate and blood pressure in the chick embryo to hypothermal stress. J. Comp. Physiol. B 155, 195–200.

Tazawa, H., Whittow, G.C., 2000. Incubation physiology. In: Whittow, G.C. (Ed.), Sturkie's Avian Physiology. Academic Press, San Diego, pp. 617–634.

Tazawa, H., Yamaguchi, S., Yamada, M., Doi, K., 1992. Embryonic heart rate of the domestic fowl (*Gallus domesticus*) in a quasiequilibrium state of altered ambient temperatures. Comp. Biochem. Physiol. 101A, 103–108.

Tazawa, H., Moriya, K., Tamura, A., Komoro, T., Akiyama, R., 2001. Ontogenetic study of thermoregulation in birds. J. Therm. Biol. 26, 281–286.

Tazawa, H., Moriya, K., Tamura, A., Akiyama, R., 2002. Low-frequency oscillation of instantaneous heart rate in newly hatched chicks. Comp. Biochem. Physiol. A 131, 797–803.

Tazawa, H., Chiba, Y., Khandoker, A.H., Dzialowski, E.M., Burggren, W.W., 2004. Early development of thermoregulatory competence in chickens: responses of heart rate and oxygen uptake to altered ambient temperatures. Avian Poult. Biol. Rev. 15, 166–176.

Yoneta, H., Fukazawa, K., Dzialowski, E.M., Burggren, W.W., Tazawa, H., 2006a. Does sequence of exposure to altered ambient temperatures affect the endothermic heart rate response of newly hatched chicks? In: Yahav, S., Tzschentke, B. (Eds.), New Insights into Fundamental Physiology and Peri-natal Adaptation of Domestic Fowl. Nottingham Univ. Press, Nottingham, pp. 15–28.

Yoneta, H., Fukuoka, S., Akiyama, R., Tazawa, H., 2006b. Early development of cholinergic heart rate control in embryos of broiler and White Leghorn chicken. In: Yahav, S., Tzschentke, B. (Eds.), New Insights into Fundamental Physiology and Peri-natal Adaptation of Domestic Fowl. Nottingham Univ. Press, Nottingham, pp. 1–14.

Zhao, R., Muehlbauer, E., Decuypere, E., Grossmann, R., 2004. Effect of genotype-nutrition on growth and somatotropic gene expression in the chicken. Gen. Comp. Endocrinol. 136, 2–11.