The Effect of Early Age Feed Restriction on Subsequent Response to High Environmental Temperatures in Female Broiler Chickens

I. Zulkifli,*[,1] M. T. Che Norma,* D. A. Israf,† and A. R. Omar†

*Department of Animal Science and †Faculty of Veterinary Medicine, Universiti Putra Malaysia, 43400 UPM Serdang, Selangor, Malaysia

ABSTRACT This study was conducted to determine whether early age feed restriction improves heat tolerance in female broiler chickens. Chicks were brooded for 3 wk and then maintained at 24 ± 1 C. On Day 0, chicks were assigned to one of four feeding regimens; each regimen was applied to four cages of chicks. The feeding regimens were 1) ad libitum feeding (ALF); 2) 40% feed restriction at 4, 5, and 6 d of age (F40); 3) 60% feed restriction at 4, 5, and 6 d of age (F60); and (4) 80% feed restriction at 4, 5, and 6 d of age (F80). From 35 to 41 d of age, all birds were exposed to 38 ± 1 C for 2 h/d. Serum concentrations of glucose were elevated by the heat challenge, but were not affected by the feeding regimen. The heat treatment resulted in hypocholesteremia among ALF and F80 chicks, whereas the concentrations increased and remained constant in the F60 and F40 birds, respectively. Subjecting chicks to F60 improved growth and survivability and reduced heterophil to lymphocyte ratios (H/L) in response to the heat treatment as compared with the ALF and F80 regimens. The survivability rate and H/L of F40 chicks were similar to those attained by chicks on other regimens. Newcastle disease antibody titer of ALF birds declined with duration of heat treatment. It is concluded that the F60 regimen is beneficial for alleviating, at least in part, the detrimental effects of heat stress in female broiler chickens.

(*Key words*: heat stress, early age feed restriction, heat tolerance, broiler chickens)

2000 Poultry Science 79:1401–1407

INTRODUCTION

Stress is often viewed as deleterious and is associated with injury, pain, disease, or death. However, there is a growing body of evidence showing that relatively mild or brief periods of stressful experiences during the neonatal period may be beneficial in preparing the body for a response to subsequent disruption of homeostasis (Zulkifli and Siegel, 1995). Although one of the earliest studies on the influences of early experience on adult adrenocortical functions was in rats (Levine, 1962), its implications in the context of veterinary science and animal agriculture have been of major interest. Gross (1983) indicated that stresses that occur early in life, while many systems of the chicks are still developing, may have long-lasting impact and could possibly modify the expression of their genetic potential.

Early age thermal conditioning has been shown to ameliorate the deleterious effects of heat stress in poultry (Arjona et al., 1988, 1990; Yahav and Hurwitz, 1996; Yahav and Plavnik, 1999). Zulkifli et al. (1994a,b) demonstrated that chicks fasted early in life had smaller increases in heterohil to lymphocyte ratios (H/L), improved resistance to marble spleen disease infection, and greater growth than those fed ad libitum in response to thermal insults later in life. Conversely, although neonatal thermal conditioning improved heat tolerance in broilers, subjecting them to concurrent feed restriction as an additional treatment failed to enhance further the ability to withstand high ambient temperatures (Yahav and Plavnik, 1999). In view of this, there is a possibility that the magnitude of stress perceived early in life may affect the level of tolerance to a subsequent stressor. To the best of our knowledge, the relationship between severity of early age stressful experience and tolerance to subsequent insults has not been documented.

It is well established that there are genetic components in response to heat stress in poultry (Smith and Oliver, 1971; Washburn, 1985; Gowe and Fairfull, 1995). Genetic differences in elicitation of improved heat tolerance through prior exposure to stress have been documented. Although repeated handling did not increase heat resistance in White Leghorns, the reverse has been observed

Received for publication December 13, 1999.
Accepted for publication June 19, 2000.
[1]To whom correspondence should be addressed: zulkifli@agri.upm.edu.my.

Abbreviation Key: ALF = ad libitum feeding; F40 = 40% feed restriction at 4, 5, and 6 d of age; F60 = 60% feed restriction at 4, 5, and 6 d of age; F80 = 80% feed restriction at 4, 5, and 6 d of age; FCR = feed conversion ratio; H/L = heterophil to lymphocyte ratio, ND = Newcastle disease.

in broilers (Bowen and Washburn, 1984). Similarly, despite the ability to induce enhanced thermotolerance in normal White Plymouth Rocks by neonatal feed restriction, the procedure had negligible impact on dwarf chicks (Zulkifli et al., 1994a). Given that early age feed restriction offers a feasible method in ameliorating heat stress-related responses in White Rocks, additional studies are needed to evaluate the benefits of such a husbandry procedure in commercial broiler chickens. The objectives of this study were to evaluate the effect of early age feed restriction on heat tolerance in female commercial broiler chickens and evaluate the relationship between magnitude of stress perceived early in life and the level of tolerance to a subsequent stressor.

MATERIALS AND METHODS

Animals, Husbandry, Environmental Temperature, and Traits Measured

One-hundred ninety-two female broiler chicks (Shaver) were obtained from a local hatchery. Upon arrival, the chicks were wing-banded, weighed, and assigned at random in groups of 12 to 16 cages of three-tiered batteries with wire floors in an environmentally controlled chamber (2.3 × 9.1 × 3.8 m). Floor space allowed was 923 cm^2 per bird. Ambient temperature on Day 0 was set at 32 ± 1 C and then gradually decreased until 24 ± 1 C was reached by Day 21. The chicks were administered live Newcastle disease (ND) vaccine[2] intraocularly on Days 7 and 21. Chicks were fed standard broiler starter (crumble form; 21% CP and 2,950 kcal ME/kg) and finisher (pellet form; 19% CP and 3,100 kcal ME/kg) diets from Day 0 to 21 and Day 22 onward, respectively. Water was available at all times, and lighting was continuous.

On Day 0, chicks were assigned to one of four feeding regimens with four cages per group. The feeding regimens were 1) ad libitum feeding (ALF); 2) 40% feed restriction at 4, 5, and 6 d of age (F40); 3) 60% feed restriction at 4, 5, and 6 d of age (F60); and 4) 80% feed restriction at 4, 5, and 6 d of age (F80). The feed restrictions were 40, 60, and 80% of the previous day's feed intake of the ad libitum group. Prior to feeding, individual BW were obtained on Days 0, 7, 14, 21, 28, 35, and 42. Weekly feed consumption was recorded, and feed conversion ratios (FCR) were determined.

The day following the feed restriction period, eight chicks from each feeding regimen were chosen randomly, and their blood (via wing vein) samples (0.3 mL) were collected in tubes containing EDTA as anticoagulant. Blood smears were prepared using May-Grünwald-Giemsa stain; heterophil and lymphocyte were counted to a total of 60 cells (Gross and Siegel, 1983). Four (one bird per cage) birds per feeding regimen were killed by cervical dislocation, and brain samples were collected for an experiment conducted concurrently with this study.

Between Day 35 and 41, all chicks (at the same time) were subjected to 2-h episodes of heat stress at 38 ± 1 C each day in the environmentally controlled chamber to evaluate their ability to withstand high ambient temperatures. The increase from 24 ± 1 C to this temperature occurred over an approximately 1.5-h period. Relative humidity was not controlled but remained below 60%. Prior to heat exposure (Day 35), eight chicks per feeding regimen were randomly chosen and bled (2.0 mL) for heterophil and lymphocyte counts, serum concentrations of glucose, cholesterol, and ND antibody titers. Feed was not removed prior to collection of blood samples because the procedure may influence birds' subsequent response to heat stress (Zulkifli and Fauzi, 1996). Blood samples for total glucose and cholesterol, and antibody titers were serum separated and stored at –20 C. Analyses for total glucose and cholesterol were conducted on an automated spectrophotometer[3] using a standard diagnostic kit. Serum samples for ND antibodies were analyzed using ELISA kit.[4] Four (one bird per cage) birds per feeding regimen were killed as described previously. Similar procedures were repeated following 4 d (Day 38) and 7 d (Day 41) of heat treatment. Mortality from Days 35 to 41, during the heat exposure, was recorded daily.

Statistical Analyses

Data were subjected to analysis of variance using the General Linear Models procedure of SAS® (SAS Institute Inc., 1991). Prior to analyses, BW and antibody titers were transformed to common logarithms. Body weight, feed intake, and FCR data were analyzed with feeding regimen as the main effect. Feeding regimen, stage of heat treatment, and their interactions were considered as the main effects for the analyses of H/L and serum concentrations of glucose, cholesterol, and ND antibody response data. When interactions between main effects were significant, comparisons were made within each experimental variable. Means were subjected to Duncan's multiple range test. Data on survivability rate were analyzed by the chi-square test. The level of significance was reported at $P \leq 0.05$.

RESULTS

Data on BW, feed intake, and FCR are presented in Table 1. The effect of early feed restriction on BW was evident by Day 7; BW of F80, F60, and F40 chicks were approximately 83, 72, and 63% of those fed ALF, respectively (Table 1). On Day 14, although F80, F60 and F40 birds had similar BW, they weighed less than their ALF counterparts. Except for F40, the F80 and F60 birds at-

[2]Nobilis ND Clone 30, Intervet International, 58300 AA Boxmeer, The Netherlands.
[3]Ultraspec® 300; Cobas-Mira, Roche Diagnostic System, CH4070 Basel, Switzerland.
[4]IDEXX Laboratory, Inc., Westbrook, ME 04029.

TABLE 1. The effect of feeding regimen[1] on BW, feed consumption, and feed conversion ratio (FCR) of broiler chickens

Parameter	Feeding regimen				Pooled SEM
	ALF	F80	F60	F40	
BW (g per bird)					
Day 0	45	46	45	46	0.29
Day 7	170[a]	141[b]	123[c]	107[d]	0.92
Day 14	372[a]	364[b]	347[b]	334[b]	2.42
Day 21	717[a]	718[a]	698[a]	664[b]	6.05
Day 28	1,168[a]	1,173[a]	1,150[ab]	1,114[b]	8.04
Day 35	1,553	1,542	1,548	1,538	10.37
Day 42	1,852[b]	1,847[b]	1,921[a]	1,828[b]	11.55
Feed consumption (g per bird)					
Days 0–6	146[a]	121[b]	104[c]	84[d]	2.0
Days 7–13	328	337	335	319	4.0
Days 14–20	572[a]	548[ab]	563[a]	526[b]	4.4
Days 21–27	807	769	806	734	10.6
Days 28–34	834	811	863	817	10.4
Days 35–41	924	888	945	895	11.3
Total	3,611[a]	3,474[ab]	3,615[a]	3,374[b]	35.3
FCR (feed/gain)					
Days 0–6	1.17[c]	1.27[bc]	1.33[ab]	1.38[a]	0.03
Days 7–13	1.62	1.51	1.50	1.41	0.03
Days 14–20	1.66	1.55	1.60	1.59	0.01
Days 21–27	1.79	1.69	1.78	1.63	0.02
Days 28–34	2.17	2.20	2.17	2.21	0.03
Days 35–41	3.09	2.91	2.53	3.09	0.12
Overall	2.00	1.93	1.93	1.89	0.08

[a–d]Means within a row-subgroup with no common letters differ significantly ($P \leq 0.05$).

[1]All chicks were fed ad libitum (ALF) from hatch to Day 3. On Days 4, 5, and 6, some chicks were subjected to F80, F60, and F40: F80 = 80% feed restriction, F60 = 60% feed restriction, and F40 = 40% feed restriction.

tained complete compensatory growth attributed to fasting by Day 21 with (AL = F80 = F60) > F40. Prior to heat treatment (Day 35), BW was not affected by feeding regimen. Following the daily 2 h of heat challenge for 7 d, F60 birds had greater BW than those on other regimens. No significant differences were noted in the BW of AL, F80, and F40 birds on Day 42.

Beginning on Day 7, except for the period from Days 14 to 20, in which F40 chicks consumed less feed than ALF and F60 chicks, feeding regimen had no significant effect on weekly feed intake. The total feed intake (from Day 0 to 41) was lower for F40 chicks than for ALF and F60 chicks. There was no significant difference between the latter two groups. The total feed intake of the F80 chicks was similar to those on other regimens. From Days 0 to 6, although ALF and F80 birds had similar FCR, the former were more feed efficient than were F60 and F40 birds. Feed conversion ratios during the heat treatment were not affected by feeding regimen. No significant differences were observed among the four feeding regimens in the overall FCR.

The mean percentages of survivability during the heat treatment are shown in Figure 1. The heat challenge resulted in high mortality among the ALF (17%) and F80 (10%) birds but not among those fed F60 (0%) and F40 (5%).

Irrespective of the 4- to 6-d of age feeding regimen, the heat treatment elevated serum concentrations of glucose, and the highest values were noted following 4 d of heat exposure (Table 2). The parameter was not affected by feeding regimen. A significant feeding regimen by stage of heat treatment interaction was observed for serum total cholesterol (Table 3). Four days of heat treatment resulted in hypocholesteremia among ALF and F80 chicks, whereas the concentrations increased and remained constant in the F60 and F40 chicks, respectively.

Interaction of feeding regimen by stage of heat exposure was significant for ND antibody titers (Table 4). Except for ALF chicks, the antibody titers of the F80,

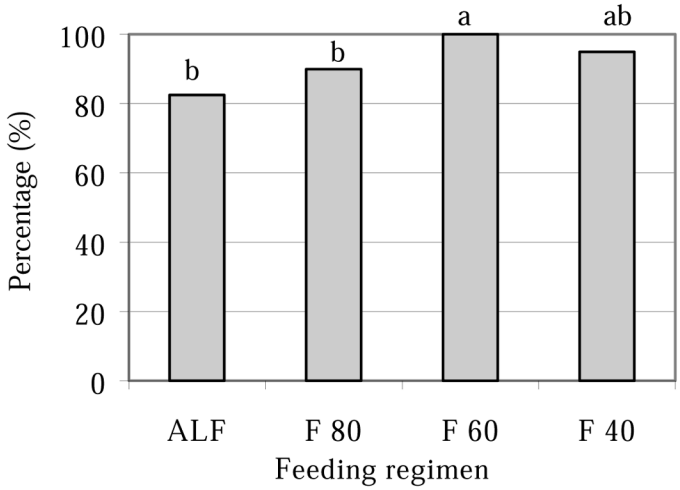

FIGURE 1. The effect of early feeding regimen on survivability rate of broiler chickens during heat treatment (Days 35 to 41). [a,b]Means with no common letters differ significantly ($P \leq 0.05$). ALF = Ad libitum feeding; F 80 = 80% feed restriction; F 60 = 60% feed restriction; F 40 = 40% feed restriction.

TABLE 2. The effect of stage of heat treatment[1] and early feeding regimen[2] on serum glucose concentrations of broiler chickens

Variable	Glucose (Mmol/L)
Stage of heat treatment	
Prior to heat	10.47[c]
4 d of heat treatment	14.88[a]
7 d of heat treatment	13.05[b]
Feeding regimen	
ALF	13.45
F80	12.45
F60	12.53
F40	12.52
Pooled SEM	0.23

[a–c]Means within a column-subgroup with no common letters differ significantly ($P \leq 0.05$).

[1]From Days 35 to 41, all chicks were exposed to 38 ± 1 C temperatures for 2 h/d.

[2]All chicks were fed ad libitum (ALF) from hatch to Day 3. On Days 4, 5, and 6, some chicks were subjected to F80, F60, and F40: F80 = 80% feed restriction, F60 = 60% feed restriction, and F40 = 40% feed restriction.

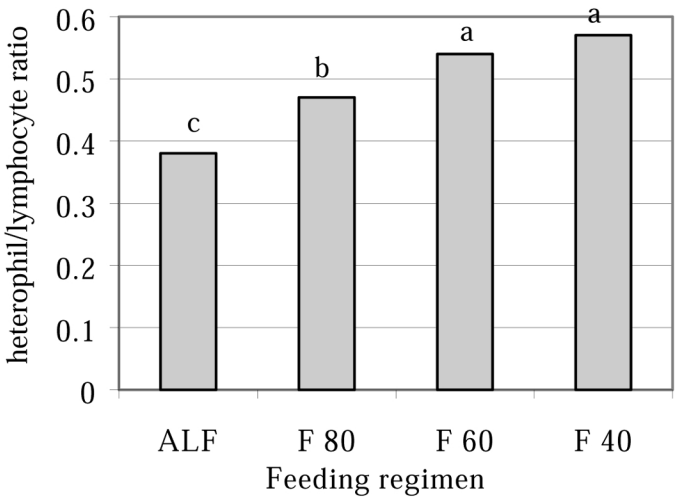

FIGURE 2. The effect of early feeding regimen on heterophil to lymphocyte ratio of broiler chickens one day following feed restriction (Day 7). [a,b]Means with no common letters differ significantly ($P \leq 0.05$). ALF = Ad libitum feeding; F 80 = 80% feed restriction; F 60 = 60% feed restriction; F 40 = 40% feed restriction.

F60, and F40 birds remained constant throughout the 7-d period of heating, which resulted in the interaction for the trait. Within the ALF group, ND antibody titers were similar and lower following 4 and 7 d of heat exposure, respectively, as compared with values attained prior to the onset of heat challenge.

The H/L response to feed restriction (Day 7) is shown in Figure 2. Following the feed restriction period, the H/L of F60 and F40 birds were higher than those of ALF and F80 birds. There was no significant difference between the former two groups. Although H/L were not affected by the early age feeding regimen prior to heat exposure (Day 35), ALF, F80, F60, and F40 birds responded differently following 4 and 7 d of heating, resulting in significant feeding regimen by stage of heat treatment interactions for the parameter (Table 5). Although the heat challenge augmented H/L response, the F60 birds had smaller increases compared with their ALF and F80 counterparts. The H/L of F40 birds were similar to those of birds in other groups throughout the heating period.

DISCUSSION

The noted elevation in H/L in response to feed restriction is not unexpected. It is well documented that fasting may elicit physiological stress response (Gross and Siegel, 1986; Katanbaf et al., 1989; Zulkifli et al., 1993, 1994a,b, 1995). It is interesting to note that on the day following feed restriction, F60 and F40 birds had similar H/L. These findings may appear unexpected in view of the assumption that the perceived magnitude of stress attributed to F40 is higher than F60. Indisputable explanations for this apparent contradiction cannot be offered. First, the sensitivity of H/L as a biological index of stress is inadequate to detect the difference in the magnitude

TABLE 3. Mean serum cholesterol concentration (Mmol/L) where stage of heat treatment[1] by early feeding regimen[2] interactions were significant

	Stage of heat treatment		
Feeding regimen	Prior to heat	4 d of heat treatment	7 d of heat treatment
ALF	6.30[a,x]	5.32[y]	4.26[z]
F80	6.04[a,x]	5.19[y]	3.96[z]
F60	4.95[b,y]	5.86[x]	4.01[z]
F40	5.42[ab,x]	4.80[x]	3.62[y]

[a,b]Means within a column-subgroup with no common letters differ significantly ($P \leq 0.05$).

[x–z]Means within a row-subgroup with no common letters differ significantly ($P \leq 0.05$).

[1]From Days 35 to 41, all chicks were exposed to 38 ± 1 C temperatures for 2 h/d.

[2]All chicks were fed ad libitum (ALF) from hatch to Day 3. On Days 4, 5, and 6, some chicks were subjected to F80, F60, and F40: F80 = 80% feed restriction, F60 = 60% feed restriction, and F40 = 40% feed restriction. Pooled SEM = 0.08.

TABLE 4. Mean Newcastle disease antibody titers[1] where stage of heat treatment[2] by early feeding regimen[3] interactions were significant

	Stage of heat treatment		
Feeding regimen	Prior to heat	4 d of heat treatment	7 d of heat treatment
ALF	3,149[a]	2,318[a,b]	1,177[b]
F80	2,834	1,640	1,647
F60	2,814	2,703	2,573
F40	2,622	2,019	1,957

[a,b]Means within a row-subgroup with no common letters differ significantly ($P \leq 0.05$).

[1]Antibody titers were measured by ELISA.

[2]From Days 35 to 41, all chicks were exposed to 38 ± 1 C temperatures for 2 h/d.

[3]All chicks were fed ad libitum (ALF) from hatch to Day 3. On Day 4, 5, and 6, some chicks were subjected to F80, F60, and F40: F80 = 80% feed restriction, F60 = 60% feed restriction, and F40 = 40% feed restriction. Pooled SEM = 177.66.

TABLE 5. Mean heterophil to lymphocyte ratios where stage of heat treatment[1] by early feeding regimen[2] interactions were significant

	Stage of heat treatment		
Feeding regimen	Prior to heat	4 d of heat treatment	7 d of heat treatment
ALF	0.35^{y}	0.66^{a,x}	0.76^{a,x}
F80	0.38^{z}	0.64^{a,y}	0.73^{a,x}
F60	0.35^{y}	0.55^{b,x}	0.57^{b,x}
F40	0.34^{y}	0.61^{ab,x}	0.66^{ab,x}

^{a,b}Means within a column-subgroup with no common letters differ significantly ($P \leq 0.05$).

$^{x-z}$Means within a row-subgroup with no common letters differ significantly ($P \leq 0.05$).

[1]From Days 35 to 41, all chicks were exposed 38 ± 1 C temperatures for 2 h/d.

[2]All chicks were fed ad libitum (ALF) from hatch to Day 3. On Days 4, 5, and 6, some chicks were subjected to F80, F60, and F40: F80 = 80% feed restriction, F60 = 60% feed restriction, and F40 = 40% feed restriction. Pooled SEM = 0.01.

of stress attributed to F60 and F40. However, this argument is weakened by the numerous reports that H/L is a reliable indicator of stress in avian species (Gross and Siegel, 1983; Maxwell, 1993). Second, and more likely, following 3 d of 40% feed restriction (i.e., the time of blood sampling for H/L), waning of the birds' response might have occurred. Evidence is accumulating to show that chickens readily habituate to fasts of moderate duration (Freeman et al., 1981; Gross and Siegel, 1986; Zulkifli et al., 1993).

The rise in total serum glucose in response to heat treatment is consistent with earlier studies (Kutlu and Forbes, 1993). These results are suggestive of elevated plasma concentrations of glucocorticoids that may increase catabolism of protein and fatty tissue through gluconeogenesis (Freeman, 1971). The observed hyperglycemia attributed to heat stress in the present study, however, is in disagreement with studies in mammalian species (Alnaimy et al., 1992). The authors attributed the decline in blood glucose concentration during heat stress to a decrease in concentration of thyroxine, which is closely associated with energy metabolism during heat exposure. Consistent with the report by Zulkifli et al. (1999), serum concentration of cholesterol declined in response to heat stress. Increase in total body water or decrease in acetate concentration, which is the primary precursor for the synthesis of cholesterol (Alnaimy et al., 1992), may account for the phenomenon.

Collectively, the data presented here suggest that the F60 chicks were more tolerant to high ambient temperatures as juveniles than were ALF, F80, and F40 chicks. Heat-induced growth retardation, mortality, and elevation of H/L were all reduced by subjecting chicks to 60% feed restriction at 4, 5, and 6 d of age. Results of this experiment concur with earlier findings concerning the possibility of evoking enhanced thermotolerance by mild feed restriction early in life (Zulkifli et al., 1994a,b). One of the objectives of our study was to evaluate whether the magnitude of stress perceived early in life influences the level of tolerance to a subsequent stressor. Although the F80 birds were feed restricted at 4, 5 and 6 d of age, there is little indication that thermotolerance can be markedly improved by such practice. In comparison with their F60 counterparts, the F80 birds had significantly higher increases in H/L, mortality rate, and weight loss in response to heat challenge. It is interesting to note that, although numerically higher, the H/L and mortality rate of the heat-stressed F40 birds were not significantly different from those of F60 birds. However, the lack of significant differences in those parameters among the ALF, F80, and F40 birds suggests that a more severe feed restriction (as compared with F60) did not enhance heat tolerance further. Hence, it appears that the magnitude of physiological stress response experienced by the chicks during the neonatal stimulation may have a profound influence on ability to withstand high ambient temperatures later in life. It is, therefore, concluded that the alleviation of effects of heat stress on growth, survivability, and leukocytic count through F60 is more profound than those through F80 and F40.

Sykes and Fataftah (1986) expressed doubts concerning the practicality of acclimating in advance by exposing chicks to controlled increases in environmental temperatures. Zulkifli et al. (1994a) indicated that subjecting chicks to early age feed restriction is more realistic under practical situations than is thermal preconditioning, such as rearing in open-sided houses where manipulation of house temperature is a problem. Yahav and Plavnik (1999), however, reported that feed restriction from 7 to 14 d was less effective than prestressing with heat in improving heat tolerance in broiler chickens. The discrepancies between these studies could stem from the differences in the protocol of early age fasting. The feed restriction practiced by Yahav and Plavnik (1999) included limiting energy intake to support 50% of normal growth rate, whereas, in the experiment reported here, birds were subjected to 60% feed restriction based on ad libitum intake, which may be less severe. Our data suggest that the severity of the early age fasting may have profound impact on the magnitude of improvement in heat tolerance later in life. Another obvious variation in the experimental design of Yahav and Plavnik (1999) and ours is the age of the chicks during the onset of fasting. It has been shown that there is a possible sensitive period for optimal neonatal stimulation, leading to improved tolerance to stress (Sapolsky, 1992; Zulkifli et al., 1994a,b).

The precise physiological mechanisms underpinning the thermoregulating effects of neonatal stimulation have yet to be determined. Some research investigating early age thermal conditioning leading to enhanced thermotolerance worked on the premise that the phenomenon is associated with a reduction in plasma triiodothyronine concentration, hemodynamic changes, increases in sensible heat loss, and efficient control of body water economy (Yahav and Hurwitz, 1996; Yahav and Plavnik, 1999). Zulkifli et al. (1994b, 1995) indicated that neonatal stimulation of the stress response with a mild stressor

without concurrent increases in the synthesis and liberation of corticosteroid might not aid an animal in responding to subsequent environmental insult. The authors concluded that corticosterone plays a role in simultaneously inducing persistent modifications of the adrenocortical function and habituation. Another possible explanation for the phenomenon is the formation of stress-elicited or heat shock proteins that may have considerable influence on resistance to both thermal and non-thermal stressors (Etches et al., 1995). However, although Wang and Edens (1998) indicated that early age heat conditioning resulted in enhanced heat shock protein expression in response to subsequent thermal challenge, Yahav et al. (1997) reported otherwise. It is uncertain whether or not neonatal fasting, leading to the acquisition of improved heat tolerance, is elicited by these physiological and cellular alterations. The actual underlying mechanisms responsible for the fasting-elicited thermotolerance await clarification.

Although the contention concerning the physiological and biochemical modifications involved in early age feed restriction and thermoregulation remain unresolved, the study reported here strengthens the notion that an animal does not always have to be preconditioned to the same stressor for adaptation to take place (Siegel, 1995; Zulkifli and Siegel, 1995). The phenomenon could be partly explained by nonspecific stress responses and acclimation (Siegel, 1995). In his review, Siegel (1995) presented a body of evidence suggesting that acclimation may have a profound impact on ameliorating nonspecific stress reactions.

It is well documented that environmental fluctuations leave an individual vulnerable to neuroendocrine alterations, thereby hindering immunity and disease defense (Thaxton, 1978; Kelley, 1985; Zulkifli, 1995). Zulkifli et al. (1994a) reported that early age fasting had a negligible effect on antibody response to sheep erythrocyte in heat-stressed White Rocks compared with those fed ad libitum. In the present study, the ND antibody values of ALF birds declined with the number of days on heat treatment, but this was not true for F80, F60, and F40 birds. Although there was a marked numerical difference in the ND antibody titers between ALF (n = 1,177) and F60 (n = 2,573) chicks, statistical analysis revealed no significant difference. These findings suggest that bird to bird variations within group were sufficient to preclude statistical significance.

In conclusion, under the conditions of these experiment, F60 appears to be beneficial in improving growth and survivability of female broiler chickens exposed to heat stress later in life.

REFERENCES

Alnaimy, A., M. Habeeb, I. Fayaz, M. Marai, and T. H. Kamal, 1992. Heat stress. Pages 22–41 *in*: Farm Animal and the Environment. C. Phillip and D. Piggins, ed. CAB International, Wallingford, UK.

Arjona, A. A., D. M. Denbow, and W. D. Weaver, 1988. Effects of heat stress early in life on mortality of broilers exposed to high environmental temperatures prior to marketing. Poultry Sci. 67:226–231.

Arjona, A. A., D. M. Denbow, and W. D. Weaver, 1990. Neonatally induced thermotolerance: Physiological responses. Comp. Biochem. Physiol. 95:393–399.

Bowen, S. J., and K. W. Washburn, 1984. Preconditioning to heat stress by nontemperature stressor. Poultry Sci. 63:917–919.

Etches, R. J., I. M. John, A.M.V. Gibbins, 1995. Behavioural, physiological, neuroendocrine and molecular responses to heat stress. Pages 31–65 *in*: Poultry Production in Hot Climates. N. J. Daghir, ed. CAB International, Wallingford, UK.

Freeman, B. M., 1971. Stress and the domestic fowl: A physiological appraisal. World's Poult. Sci. J. 27:263–275.

Freeman, B. M., A.C.C. Manning, and I. H. Flack, 1981. The effects of restricted feeding on adrenal cortical activity in the immature domestic fowl. Br. Poult. Sci. 22:295–303.

Gowe, R. S., and R. W. Fairfull, 1995. Breeding for resistance to heat stress. Pages 11–29 *in*: Poultry Production in Hot Climates. N. J. Daghir, ed. CAB International, Wallingford, UK.

Gross, W. B., 1983. Chicken-environment interactions. Pages 329–337 *in*: Ethics and Animals. H. B. Miller and W. H. Williams, ed. Humana Press, Clifton, NJ.

Gross, W. B., and H. S. Siegel, 1983. Evaluation of heterophil/lymphocyte as a measurement of stress in chickens. Avian Dis. 27:972–979.

Gross, W. B., and P. B. Siegel, 1986. Effect of initial and second period of fasting on heterophil and lymphocyte ratios and body weight. Avian Dis. 30:345–346.

Katanbaf, M. N., E. A. Dunnington, and P. B. Siegel., 1989. Restricted feeding in early and late feathering chickens. 1. Growth and physiological responses. Poultry Sci. 68:344–351.

Kelley, K. W., 1985. Immunological consequences of changing environmental stimuli. Pages 193–223 *in*: Animal Stress. G. P. Moberg, ed. Waverly Press, Inc., Baltimore, MD.

Kutlu, H. R., and J. M. Forbes, 1993. Changes in growth and blood parameters in heat-stressed broiler chicks in response to dietary ascorbic acid. Livestock Prod. Sci. 36:335–350.

Levine, S., 1962. Plasma-free corticosteroid response to electric shock in rats stimulated in infancy. Science 135:795–796.

Maxwell, M. H., 1993. Avian blood leucocyte response to stress. World's Poult. Sci. J. 49:34–43.

Sapolsky, R. M., 1992. Stress, the Aging Brain and the Mechanism of Neuron Death. MIT Press, Cambridge, MA.

SAS® Institute, 1991. SAS/STAT User's Guide. Release 6.03 ed. SAS Institute Inc, Cary, NC.

Siegel, H. S., 1995. Stress, strains and resistance. Br. Poult. Sci. 36:3–22.

Smith, A. J., and L. Oliver, 1971. Some physiological effects of high temperature on the laying hen. Poultry Sci. 50:912–916.

Sykes, A. H., and A.R.A. Fataftah, 1986. Acclimatization of the fowl to intermittent acute heat stress. Br. Poult. Sci. 27:289–300.

Thaxton, P., 1978. Influence of temperature on the immune response of birds. Poultry Sci. 51:2617–2623.

Wang, S., and F. W. Edens, 1998. Heat conditioning induces heat shock proteins in broiler chickens and turkey poults. Poultry Sci. 77:1636–1645.

Washburn, K. W., 1985. Breeding of poultry in hot and cold environments. Pages 111–122 *in*: Stress Physiology in Livestock. Vol. 3—Poultry. M. K. Yousef, ed. CRC Publications, Boca Raton, FL.

Yahav, S., and S. Hurtwitz, 1996. Induction of thermotolerance in male broiler chickens by temperature conditioning at an early age. Poultry Sci. 75:402–406.

Yahav, S., and I. Plavnik, 1999. Effect of early-age thermal conditioning and food restriction on performance and thermotolerance of male broiler chickens. Br. Poult. Sci. 40:120–126.

Yahav, S., A. Shamay, G. Horev, D. Bar-ilan, O. Genina, and M. Friedman-Einat, 1997. Effect of acquisition of improved thermotolerance on the induction of heat shock proteins in broiler chickens. Poultry Sci. 76:1428–1434.

Zulkifli, I., 1995. Stress, immunity and disease resistance in poultry. A review. J. Vet. Malay. 7:1–6.

Zulkifli, I., R. T. Dass, and M. T. Che Norma, 1999. Acute heat stress effects on physiology and fear-related behaviour in red jungle fowl and domestic fowl. Can. J. Anim. Sci. 79:165–170.

Zulkifli, I., E. A. Dunnington, W. B. Gross and P. B. Siegel, 1994a. Food restriction early or later in life and its effect on adaptability, disease resistance, and immunocompetence of heat stressed dwarf and nondwarf chickens. Br. Poult. Sci. 35:203–214.

Zulkifli, I., E. A. Dunnington, W. B. Gross, and P. B. Siegel, 1994b. Inhibition of adrenal stroidogenesis, food restriction and acclimation to high ambient temperatures in chickens. Br. Poult. Sci. 35:417–426.

Zulkifli, I., E. A. Dunnington, W. B. Gross, A. S. Larsen, A. Martin, and P. B. Siegel, 1993. Responses of dwarf and normal chickens to feed restriction, *Eimeria tenella* challenge, and sheep red blood cell antigen. Poultry Sci. 72:1630–1640.

Zulkifli, I., and A. M. Fauzi, 1996. Behaviour and performance of fasted broilers under high temperature and humidity. J. Vet. Malay. 8(1):1–6.

Zulkifli, I., and P. B. Siegel, 1995. Is there a positive side to stress? World's Poult. Sci. J. 51:63–76.

Zulkifli, I., H. S. Siegel, M. M. Mashaly, E. A. Dunnington, and P. B. Siegel, 1995. Inhibition of adrenal steroidogenesis, neonatal feed restriction and pituitary-adrenal axis response to subsequent fasting. Gen. Comp. Endocrinol. 97:49–56.