

PRODUCTIVE AND PHYSIOLOGICAL ADAPTIVE RESPONSES OF ETHIOPIAN NAKED-NECK CHICKENS AND THEIR F₁ CROSSES WITH COMMERCIAL CHICKEN BREEDS TO HIGH ENVIRONMENTAL TEMPERATURE

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ABSTRACT: The objective of this study was to evaluate the effects of the interaction between chicken genotypes (Naked-neck, Na, from Ethiopia; New Hampshire, NH; Lohmann White, LW; and F₁ crosses of Na males with females of NH [Na×NH] and LW [Na×LW]) and ambient temperatures (normal and high) on physiological indicators and performance traits. Two-hundred forty female chickens were assigned to a completely randomized design of 2 × 5 factorial arrangements (2 temperatures and 5 genotypes). Eggs were collected daily while feed intake was determined at 28-d intervals and egg shell thickness at 4 age points. Corticosterone (CS) and 3,5,3'-triiodothyronine (T₃) levels were determined from 480 blood samples taken at 4 age points. Commercial hens reared at high temperature showed significant (p<0.05) performance reductions in egg production (33%), feed intake (15%) and shell thickness (24.3%). The effect of heat stress on T₃ levels was significant (p<0.001) and consistent across heat-stressed genotypes resulting in an overall reduction of 29% compared with those reared at normal temperature. Moreover, significant (p<0.05) differences in plasma T₃ levels were observed between heat-stressed genotypes. Although the CS levels uniformly increased due to heat stress, the response of genotypes with advancing age was inconsistent. In conclusion, the Na×LW crosses at high temperature outperformed other genotypes and thus, appeared to be suitable genetic combinations. The Na chickens and their F₁ crosses demonstrated reduced thyroid gland activity suggesting improved thermo-tolerance to long-term heat-exposure. The present findings suggest that levels of T₃ hormone might be considered as reliable indicator of long-term heat stress in chickens.

Key words/phrases: 3,5,3'-triiodothyronine, corticosterone, F₁ crosses, heat stress, Naked-neck chicken

INTRODUCTION

Poultry is affordable animal protein and promising livestock sub-sector for poverty alleviation in developing countries. However, the productivity of poultry in tropical and subtropical countries is affected by a number of factors. Stress due to high environmental temperature is widely recognised as one of the primary problems of poultry production in tropical and subtropical climates where farmers cannot afford costly artificial control of heat stress in poultry houses (Maak *et al.*, 2003; Cahaner *et al.*, 2008). Stress responses are considered to be essentially adaptive or protective and thus should prevent or minimize detrimental effects of the stressor

that was imposed upon the animal. Studying the response of chickens to heat stress revealed large variations in their reaction to heat as evaluated by blood composition and behaviour.

With the rapid development of the poultry industry worldwide, especially in developing countries characterized by hot tropical climates, importation of temperate type-high performance stocks to hot regions is on the rise. Nevertheless, the use of unsuitable genotypes in hot regions has been resulting in large economic losses due to depression in general performances and higher mortality (Yalcin *et al.*, 1997). The depression in performance cannot be fully compensated by management in developing countries where limited capital is available to reduce the heat load

in chicken houses (Cahaner and Leenstra, 1992). Thus, to achieve further improvements in the poultry industry of developing countries, breeding programs need to identify chicken genotypes of temperate origin that are appropriate for the development of suitable chicken breeds in the tropical environments.

Various heat-induced responses to environmental stressors have been used as indicator of identifying heat-tolerant animals of different genetic backgrounds. Among others, the levels of corticosterone (CS) and 3,5,3'-triiodothyronine (T₃) have been considered as reliable indicators of heat stress responses in farm animals (Siegel, 1995; Bogin *et al.*, 1996; Collin *et al.*, 2005). The adrenocorticotrophic hormone stimulates the adrenal cortex, which in turn releases corticosteroids, primarily CS in birds. Since increased levels of circulating CS have been observed under various stress situations (Davis *et al.*, 2000; Piestun *et al.*, 2008), the response to heat exposure is considered primarily as a reaction to stress. Heat stress stimulates the release of CS from the adrenal glands and increases plasma concentrations of CS in chickens (Edens and Siegel, 1975; Zulkifli *et al.*, 2009).

The thyroid hormones have been known to be involved in the control of thermoregulation in birds and mammals (Collin *et al.*, 2005). Warm-blooded animals respond to increasing ambient temperature by decreasing thyroid hormone secretion rate as ambient temperature increases, and vice versa (Silva, 2003). However, the decrease in T₃ concentration in response to heat stress may vary depending upon the duration of heat exposure, type of breed and age of the birds (Sandercock *et al.*, 2006; Tao *et al.*, 2006; Chiang *et al.*, 2008). This study was thus designed to investigate the productive and physiological adaptive responses of LW and NH commercial chicken breeds and their F₁ crosses with Ethiopian Naked-neck chicken to long-term heat stress.

MATERIALS AND METHODS

Experimental animals and their management

A total of 240 female chickens were randomly assigned to a completely randomized design in a 5 × 2 factorial arrangement consisting of five

genetic groups (Naked-neck, Na, from Ethiopia; Lohmann White, LW; New Hampshire, NH and their F₁ crosses [Na×LW and Na×NH]) and two ambient temperatures (high = 30–32°C; normal = 18–20°C). Chickens reared at normal and high ambient temperatures were considered as control and experimental groups, respectively. Both LW and NH breeds were used as a maternal line whereas the local Na was used as a paternal line to produce the F₁ crosses using artificial insemination. The birds were hatched at the same time and female chicks were separated from males by auto-sexing method through examining the relative length of the primary feathers of the wing, with the females carrying genes for fast feathering and the males carrying genes for slow feathering, having long and short primary feathers, respectively.

Twenty-four female chicks from each genotype were randomly assigned either to normal or to high ambient temperatures. The experimental chickens were raised on concrete floor pens covered with appropriate bedding materials during the brooding (0–8 weeks of age) and growing (9–20 weeks of age) periods. The growing pullets were then transferred to individual layer cages, each with a dimension of 1000 cm², at the end of the 20 weeks period. The temperature in control and in experimental houses was thermo-regulated. Ambient temperature and relative humidity of the pen were measured at 2 hours interval using a Tinytalk™ II Data Logger device (UK). Relative humidity could not be controlled but was monitored continuously and ranged from 45 to 70% and 60 to 80% in the experimental and control houses, respectively. The hens were kept under 12 hours light program, which corresponds to the natural conditions in the tropics. The management practices in experimental and control groups were essentially the same.

During the brooding and growing periods, the experimental birds had *ad libitum* access to commercial starters and growers rations shown in Table 1, respectively, and were offered adequate clean water all the times. Starting from an age of 21 weeks, they were placed on commercial layer diets in an individual cage, fed *ad libitum* (4 hens/feed pan) and supplied with individual nipple drinkers. Eggs were collected once daily. Egg weight and feed intake were determined at 28 days intervals. Egg production

was then calculated using standard methods. Shell quality traits were determined in all birds at 27, 43, 55 and 68 weeks of age. To this effect, eggs that were laid within 24 hours at each age point were used for the eggshell quality assessment. Mortality was recorded as it occurred.

Blood sampling procedures

Blood samples (2–3 ml) were collected from 12 randomly selected birds of each genotype and ambient temperature measured at 22, 38, 51 and 65 weeks age (12 birds $\times$ 2 ambient temperatures $\times$ 5 genotypes $\times$ 4 age points = 480 samples). Blood samples were taken by a qualified veterinarian from the wing vein of the bird using disposable syringes and directly collected into ethylene-diamine-tetra acetic acid (EDTA) coated test tubes. Blood was taken in the morning between 8.00 and 10.00 a.m., and the time needed between handling of each chicken and bleeding was less than one minute. Collected blood was centrifuged and plasma was stored at -20°C until further processing.

Table 1. Nutrient composition of commercial feed used for chicks, pullets and layers.

Nutrients	Chicks	Pullets	Layers
Metabolizable Energy (MJ/kg DM)	11.4	11.4	11.4
Crude protein (%)	18.0	15.2	17.4
Methionine (%)	0.35	0.30	0.37
Crude ash (%)	7.3	6.5	12.5
Crude fibre (%)	5.0	5.5	5.3
Crude fat (%)	4.0	3.5	7.0
Calcium (%)	1.0	0.9	3.5
Phosphorus (%)	0.7	0.6	0.6
Sodium (%)	0.12	0.12	0.12
Vitamin A (IU/kg)	9000	9000	12000
Vitamin D ₃ (IU/kg)	1500	1500	2500
Vitamin E (mg/kg)	15	15	20

Assessment of hormones levels

The corticosterone (CS) level was assayed with Radio immuno assay (RIA1364; DRG, Marburg, Germany). Total plasma 3,5,3'-triiodothyronine (T₃) was determined with an ELISA test (EIA1780, DRG, Marburg, Germany). In this procedure, a micro plate reader capable of readings at 450 nm wavelength was used. Assaying was performed within four weeks of blood collection. Both analyses (corticosterone and T₃ levels) were

performed essentially as described in the manufacturer's manuals. Moreover, each sample was prepared in duplicate to enhance precision.

Statistical analysis

The experiment was conducted as a completely randomized factorial 2 $\times$ 5 design that consisted of two ambient temperatures (high and normal) and five genotypes (Na, LW, NH, Na $\times$ LW and Na $\times$ NH). The samplings at the four different animal ages for hormone and eggshell quality analysis were treated as replications. Data analysis was done with the SAS PROC GLM procedures (SAS, 2002) with the model including the main effects of genotype and ambient temperature and their interactions. Comparisons of multiple means were made by using Duncan's Multiple Range Test. All statements of statistical differences were based on a significance level of $p < 0.05$ unless noted otherwise.

RESULTS

Performance traits

Among the genotypes exposed to heat stress, the overall mortality rate in Na, LW, NH and Na $\times$ NH was 4.2, 13.9, 4.2 and 4.2%, respectively. No mortality was observed in heat-exposed Na $\times$ LW genotype. In chickens kept at normal temperature, chicken death was observed only in Na $\times$ LW and LW genotypes with mortality rates of 4.2 and 8.3%, respectively.

Table 2 presents Least-square means of performance traits in Naked-neck and commercial layer hens. As shown in the table, most production parameters were severely affected by heat stress with significant genotype and temperature interactions. Age at first egg of F₁ crosses and commercial layers were significantly earlier than that of local Na chickens. Among heat-exposed genotypes, the average age at first egg was significantly earlier in LW and Na $\times$ LW compared with NH and Na $\times$ NH genotypes. The age at first egg was still shorter by three days for LW breed than its F₁ crosses. However, age at first egg for Na $\times$ NH cross was significantly earlier by three days than the NH breed.

Table 2. Least-square means of performance traits in Naked-neck and commercial layer hens with their F₁ crosses at normal and high ambient temperatures (N = 240).

Temperature (T) Genotype (G)	Normal (18–20°C)					High (30–32°C)					Pooled SEM	Significance		
	Na	LW	NH	Na×L W	Na×NH	Na	LW	H	Na×LW	Na×NH		T	G	T× G
No. of birds per treatment	24	24	24	24	24	24	24	24	24	24				
Age at first egg, d	173 ^a	154 ^d	162 ^b	156 ^{cd}	160 ^{bc}	166 ^a	151 ^d	162 ^b	154 ^d	159 ^c	1.337	**	***	NS
Body weight, 68 wks, kg	1.27 ^c	1.59 ^b	2.03 ^a	1.58 ^b	1.72 ^b	1.23 ^b	1.55 ^a	1.69 ^a	1.63 ^a	1.64 ^a	0.052	***	**	**
Egg production, %	39.2 ^c	85.3 ^a	73.5 ^b	66.8 ^b	67.6 ^b	38.4 ^c	76.1 ^a	63.6 ^b	66.9 ^{ab}	63.0 ^b	0.002	**	***	NS
Feed intake, g/d/hen	77.0 ^c	120 ^a	116 ^a	101 ^b	105 ^b	63.3 ^c	98.8 ^a	89.7 ^b	88.8 ^b	87.3 ^b	0.002	***	***	***

Note: Means between genotypes within each ambient temperature having different letters are significantly different ($p < 0.05$). Na, Naked-neck (from Ethiopia); LW, Lohmann White; NH, New Hampshire; Na×LW, F₁ crosses of Na (males) and LW (females); Na×NH, F₁ crosses of Na (males) and NH (females); *, $p < 0.05$; **, $p < 0.01$; ***, $p < 0.001$; NS, Not significant; SEM, Standard error of mean.

The body weight at 68 weeks of age was similar among heat-exposed commercial layers and their F₁ crosses. Compared with control birds reared at normal temperature, the body weight of Na×LW exposed to high temperature increased by +3.2%. The indigenous Na was significantly inferior in body weight at both ambient temperatures compared with the other genotypes.

As shown in Table 2, feed consumption and egg production were significantly affected by heat stress. The effect was more pronounced in commercial layer hens than for Na and their F₁ crosses. The average decline in feed consumption was highest in NH and lowest in Na×LW genotypes but intermediate in Na, LW and Na×NH genotypes. The effect of heat stress on egg production was most severe in commercial layers (-12.2% reduction), least severe in the Na (-2.04%) and intermediate in Na×NH genotypes (-6.8%). However, egg production in heat-stressed Na×LW genotype increased by 0.15% compared

to those kept at normal temperature. This indicated that the impact of heat stress on egg production was considerably larger in commercial layers than on the Na×LW genotype.

Table 3 presents, genotype and environment effect on shell thickness which were highly significant at all age points investigated. At normal ambient temperature, the overall shell thickness values were similar between Na, LW, NH and Na×LW genotypes while the Na×NH genotype had the lowest values which differed significantly from LW and Na×LW. At high ambient temperature, the shell thickness in the Na×LW genotype was consistently better than those of the other genotypes throughout the entire experiment. Accordingly, the overall shell thickness value in the Na×LW genotype was significantly higher than in the other genotypes. In general, the overall shell quality of the heat-exposed birds was significantly higher in F₁ crosses and Na than in the commercial layers.

Table 3. Effects of genotype and environment interactions on egg shell thickness (μm) at 27, 43, 55 and 68 weeks of birds' age (N = 240/age group).

Ambient temperature (T)	Genotype (G)	Bird's age (weeks)				Overall mean
		27	43	55	68	
Normal	Na	378 ^b	383 ^a	359 ^b	367 ^a	375 ^{ab}
	LW	399 ^a	375 ^{ab}	367 ^{ab}	373 ^a	379 ^a
	NH	386 ^{ab}	384 ^a	367 ^{ab}	367 ^a	376 ^{ab}
	Na×LW	387 ^{ab}	378 ^{ab}	377 ^a	372 ^a	378 ^a
	Na×NH	382 ^b	372 ^b	352 ^b	369 ^a	369 ^b
High	Na	358 ^b	352 ^c	348 ^a	345 ^{ab}	352 ^b
	LW	357 ^b	349 ^c	321 ^b	327 ^b	340 ^c
	NH	372 ^a	342 ^c	330 ^b	305 ^c	337 ^c
	Na×LW	385 ^a	381 ^a	362 ^a	356 ^a	371 ^a
	Na×NH	374 ^a	365 ^b	352 ^a	345 ^{ab}	359 ^b
Pooled SEM		4.85	3.81	5.85	6.27	2.91
Sources of variations		Significance levels				
T		***	***	***	***	***
G		**	***	***	***	***
T × G		***	***	***	***	***

Note: Symbols and abbreviations as in Table 2 above.

Physiological responses of plasma 3,5,3'-triiodothyronine (T_3) levels

Table 4 presents levels of plasma corticosterone and 3,5,3'-triiodothyronine in five genotypes. As presented in the table, the interaction between genotype and ambient temperature in T_3 levels was highly significant ($p < 0.001$) at all ages.

The effect of heat stress on T_3 levels was highly significant ($p < 0.001$) and consistent across all genetic groups resulting in a general depression

of about 29% compared with controls (Fig. 1). As shown in Figure 2, the T_3 level slightly increased between 22 and 38 weeks of age in Na, Na×LW and LW genotypes, whereas it decreased in NH and Na×NH genotypes. At 51 weeks of age, the T_3 level sharply declined and remained constant thereafter across all genotypes, indicating a reduced function of the thyroid gland at the later ages.

Table 4. Levels of plasma corticosterone and 3,5,3'-triiodothyronine in five genotypes (Na, LW, NH, Na×LW, Na×NH) kept at normal and high ambient temperatures.

Temperature/Age (wks)	Corticosterone (ng/ml)				3,5,3'-triiodothyronine (nmol/l)			
	22	38	51	65	22	38	51	65
Normal	3.71 ^a	4.21 ^b	3.60 ^b	3.51 ^a	6.19 ^a	6.11 ^a	3.89 ^a	3.30 ^a
High	3.88 ^a	5.02 ^a	4.47 ^a	3.93 ^a	4.75 ^b	4.57 ^b	2.48 ^b	2.27 ^b
Change (%) [#]	4.38	19.2	24.2	12.7	-23.3	-25.2	-36.3	-31.1
Pooled SEM	0.18	0.23	0.20	0.02	0.07	0.04	0.05	0.05
Sources of variations	Significance levels							
Temperature (T)	NS	**	***	*	***	***	***	***
Genotype (G)	***	***	***	***	*	***	***	***
T × G	NS	**	***	***	***	***	***	***

Note: Means between ambient temperatures within each age having different letters are significant ($p < 0.05$). *, $p < 0.05$; **, $p < 0.01$; ***, $p < 0.001$; NS, Not significant; SEM, Standard error of mean; [#]Change to normal temperature (%) = (High-Normal)/(Normal) * 100.

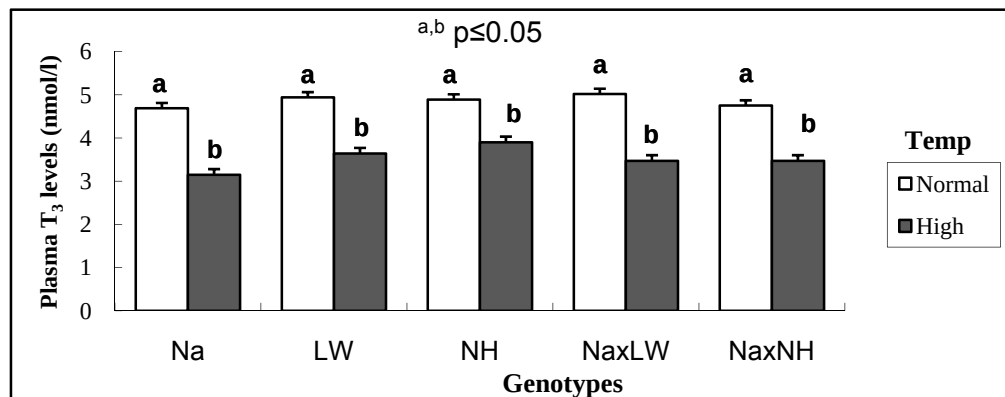


Fig. 1. Plasma levels of T_3 in Na, commercial breeds and F_1 crosses kept in normal and high ambient temperatures. (Bars indicate standard errors of the mean). Na, Naked-neck (from Ethiopia); LW, Lohmann White; NH, New Hampshire; Na×LW, F_1 crosses of Na (males) and LW (females); Na×NH, F_1 crosses of Na (males) and NH (females). Normal, control group reared at 18–20°C; High, experimental group reared at 18–20°C.

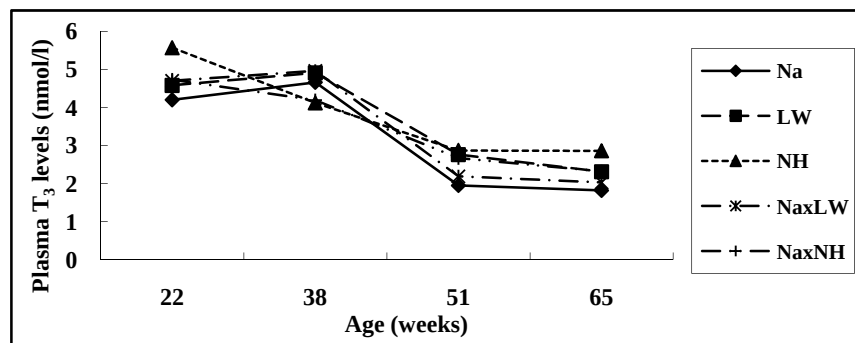


Fig. 2. Age dependent changes in plasma T_3 levels in heat-stressed genotypes. (Abbreviations as in Figure 1 above).

Physiological responses of plasma corticosterone (CS) levels

As presented in Table 4, the effect of genotype on CS levels was highly significant at all times measured, whereas that of temperature was significant at 38, 51 and 65 weeks but not at 22 weeks of age. In general, the CS level significantly increased by about 15% in heat-stressed hens compared with those of chickens reared at the normal temperature (Table 4). Nevertheless, the magnitude of heat stress on the response of CS level was inconsistent in different genotypes in which the highest level was observed in Na and Na×NH genotypes and the lowest in LW and Na×LW (Fig. 3). In heat-stressed genotypes, the overall mean values of the CS concentration at 22 weeks of age slightly decreased compared with those of birds reared at the control temperature. However, compared with control genetic groups, the overall mean values of CS concentration increased in heat-exposed genotypes by 19%, 24% and 12% at 38, 51 and 65 weeks of age, respectively.

The level of CS concentration with increasing age was similar at both ambient temperatures. The maximum CS concentration was observed at 38 weeks age and then declined sharply at 51 and thereafter slightly until 65 weeks age.

DISCUSSION

Performance traits

The general depression in performance traits (body weight, feed consumption, egg production and shell thickness) across heat-stressed genotypes is consistent with previous findings (Scott and Balnave, 1988; Mashaly *et al.*, 2004; Franco-Jimenez *et al.*, 2007). In the Na×LW genotype, the body weight was 3.2% higher at 68 weeks of age in the heat-stressed group than in the control group. This result suggests that this F₁ cross combination could be physiologically more heat-tolerant and stable than the Na and Na×LH genotypes as well as LW and NH chicken breeds. Moreover, earlier age at first egg in heat-exposed genotypes suggested improvement in the production performance.

The reduction in feed consumption in response to heat stress confirms earlier studies (Mashaly *et al.*, 2004; Lu *et al.*, 2007). Heat stress not only reduces feed intake but has been reported to also reduce digestibility of different components of the diet (Bonnet *et al.*, 1997). Furthermore, it has been reported that exposure to high temperature decreased plasma protein concentration (Zhou *et al.*, 1998) and plasma calcium concentration (Mahmoud *et al.*, 1996), both of which are required for egg formation.

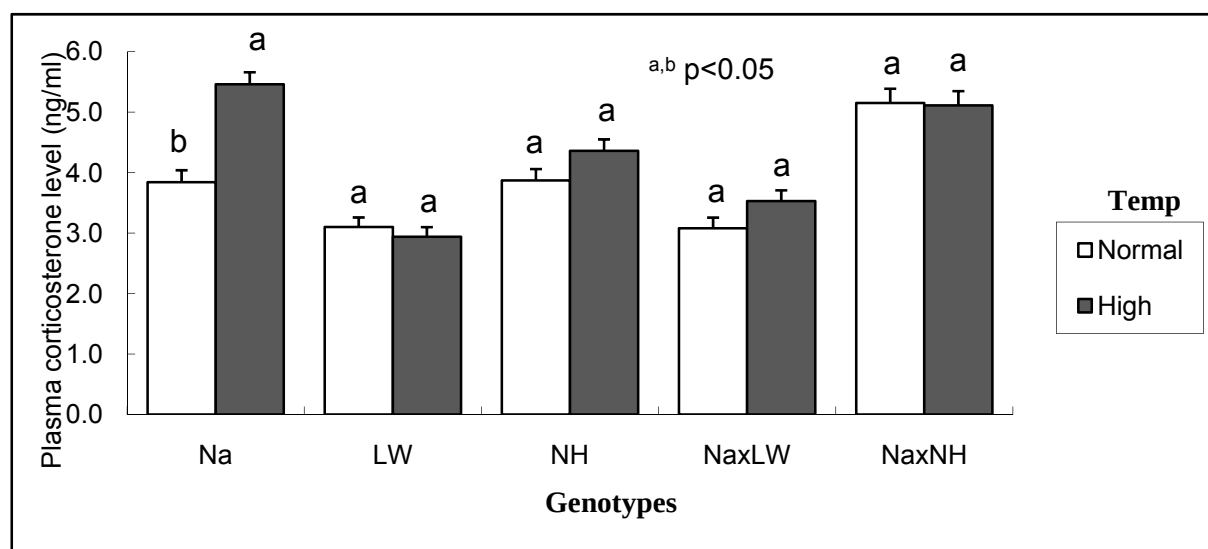


Fig. 3. Concentration of plasma corticosterone in five genetic groups reared at normal and high ambient temperatures (Bars indicate standard errors of the mean). Na, Naked-neck (from Ethiopia); LW, Lohmann White; NH, New Hampshire; Na×LW, F₁ crosses of Na (males) and LW (females); Na×NH, F₁ crosses of Na (males) and NH (females); Normal, control group reared at 18–20°C; High, experimental group reared at 18–20°C.

Plasma 3,5,3'-tri-iodothyronine (T_3)

The general reduction in T_3 level in all heat-stressed genotypes is consistent with previous reports (Silva, 2003; Gharib *et al.*, 2008). Mitchell and Carlisle (1992), and Geraert *et al.* (1996) found a sharp decline of plasma T_3 in broiler chickens reared at ambient temperatures of 35 and 32°C, respectively. As observed in the present study, the main consequence of the heat stress on animal productivity is related to a decrease in feed intake. The lower feed intake together with a decrease in blood circulating thyroid hormone levels determine lower metabolic and thermogenic rates, which explain the decrease of animal productivity during acclimatization to chronic stressful heat conditions.

The presence of significant interactions between genotypes and temperature at all age points emphasize that thermal stress influenced all studied genetic groups in different ways. Accordingly, the low T_3 concentration observed in the local Na genotype (Fig. 1) suggests improved adaptability to long-term heat-exposure due to reduced feather coverage and relative body size. Reduced feather coverage should improve and enhance heat dissipation and consequently alleviate the effects of heat on chickens reared in hot climates (Ajang *et al.*, 1993). Moreover, it has been documented that genotypes with small body size demonstrated better heat-tolerance to stressful environments (Zeman *et al.*, 1996). This may further suggest that the thyroid gland in small body-sized chickens produces little T_3 , which is beneficial for better adaptability in hot environments.

On the other hand, both commercial genotypes (LW and NH) were less effective in reducing plasma T_3 with increasing temperature, which might explain their greater difficulties in coping with long-term heat challenges, as reflected by their significant reduction in performance traits and increased mortalities particularly in LW breed, which had a mortality of 13.9%. A poor resistance to heat stress in commercial layer hens may be attributable to a reduced ability to lose heat efficiently (MacLeod and Hocking, 1993) or inappropriately increased heat production during exposure to high thermal loads (Sandercock *et al.*, 1995). Tolerance of short- or long-term elevated thermal loads is greater in unimproved local chicken breeds than commercial intensively

selected broiler or layers lines (Berrong and Washburn, 1998).

In agreement with the findings of Davis *et al.* (2000), the concentrations of circulating T_3 in the current study varied with respect to the age and egg production cycle of the hens. Plasma T_3 increased to its highest level during peak and mid egg production periods and then, declined until the end of the experiment (68 weeks of age). Lien and Siopes (1993) observed a similar response in turkeys when T_3 peaked during the early onset of lay and then steadily declined during the remaining egg production cycle. Thus, increases in T_3 during the first phase of egg production in laying hens are most likely related to adaptation to changes in metabolic demands caused by physiological stress. It could be thus speculated that basal metabolic rate might have been augmented to meet the increased demand for high egg production during this phase. As discussed above, the highly productive commercial layer hens in the current study were very similar in their endocrine profiles but differed (not significantly) from the local Na chickens. The local Na chickens were characterized by lower plasma T_3 across all ages (except at 65 weeks age) compared with commercial layer hens. Contrary to the present finding, Gonzales *et al.* (1999) reported higher T_3 levels in Naked-neck male broiler chickens at similar ages.

Plasma corticosterone (CS)

The increased level of CS in heat-exposed chickens in the current study is consistent with previous findings (Edens and Siegel, 1975; Bowen and Washburn, 1984; Davis *et al.*, 2000; Piestun *et al.*, 2008). Changes in hormonal status, particularly in CS, may have a considerable influence on responses to heat-exposure (Siegel, 1980). In the literature, the effect of heat stress on CS concentration has not been consistent. McFarlane and Curtis (1989) reported that exposing 7 days-old chicks to environmental heat stress for 7 days increased H/L ratio but not CS. Edens and Siegel (1975) indicated that increases in CS attributable to heat stress were maintained for only 70 minutes. On the contrary, Ben-Nathan *et al.* (1976) found an increased level of CS in chickens exposed to constant chronic heat stress (32.2°C) compared with those kept in control temperature (21°C).

The higher concentration of CS observed in the present study in the Na genotype may agree with the results of Vleck (1993), who reported increased plasma CS concentrations in species of wild birds of arid zones that experienced drought conditions. Various chicken breeds possess different levels of CS in the blood plasma and respond differently for the same stress. This has been demonstrated in the present study due to the presence of significant interaction between genotypes and temperature, which emphasizes that thermal stress, influenced all genotypes differently. This emphasises the role of the lower thyroid hormone concentrations in the acclimatization process by reducing metabolic rate. According to Burgess (1988), the dwarf chicken lines have a lower plasma CS level with a decreased response to acute heat stress compared with normal-sized chicken lines. This difference in thermoregulation ability of heat production or heat loss may be under genetic control.

The level of CS in the current study was affected by the age, which was related to the egg production cycle. Plasma CS considerably increased during peak egg production between 22 and 38 weeks of age and declined thereafter. The higher CS level during the peak egg production phase could be interpreted as periods of physiological stress. As CS is a gluconeogenic hormone to produce glucose from endogenous sources, usually from protein, (Davis *et al.*, 2000) and elevated CS is correlated to its metabolic effects to provide glucose and energy for peak egg production.

The CS level is closely correlated with resistance to diseases. Selection of chickens for high CS level resulted in increased resistance against bacterial diseases as well as internal and external parasites (Hartmann, 1983). On the contrary, lines selected for low CS level showed improved resistance against viral diseases associated with increased antibody production and effective immunity (Gross and Siegel, 1983) and improved feed efficiency and reproduction traits (Gross and Siegel, 1986).

Increased circulating glucocorticoid levels are known to result in gluconeogenesis with a resultant increase in circulating concentrations of glucose and heterophil/lymphocyte (H/L) ratio (Siegel, 1971). Elevated blood levels of CS caused increased energy levels by acting on intermediary metabolism of carbohydrates, protein, and fats (Olanrewaju *et al.*, 2006). CS along with other

blood-borne physiological variables is associated with ACTH-mediated gluconeogenesis from labile protein as indicated by an increase in non-protein nitrogen concomitant with increased excretory uric acid level (Siegel and van Kampen, 1984). The decline in CS level with increasing age is in accordance with Gould and Siegel (1985). With increasing age, the difference between experimental and control groups in CS level became narrower, suggesting acclimation of investigated chicken genotypes to chronic heat-exposure over time.

CONCLUSIONS

The present study clearly showed that the Naked-neck chickens and their F₁ crosses with Lohmann White and New Hampshire were much better in heat-tolerance than high performing commercial layer breeds. Thus, results of this study suggest that significant interactions between genotype and environment on most performance traits can be expected when importing commercial layer breeds to tropical climates, which potentially may result in large economic losses as observed in Lohmann White breed with the highest mortality rate. Among both F₁ crosses, the Lohmann White with local Naked-neck crossbred demonstrated the highest heterosis effect with outstanding heat-tolerance, which suggests the best genetic combination of both genotypes for tropical environment. Since responses of plasma T₃ levels were consistent in all heat-exposed genotypes, this hormone might be considered as reliable indicator of long-term heat stress in layer chickens.

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