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Comparative Ability to Tolerate Heat Between Thai Indigenous Chickens, Thai Indigenous Chickens Crossbred and Broilers by Using Heterophil/Lymphocyte Ratio

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Abstract: The effects of high environmental temperature on the heterophil/lymphocyte ratio were determined for a comparison of the ability to tolerate heat between Thai indigenous chickens, crossbred Thai indigenous chickens and broilers. One kilogram of the representative males and females of each of the three breeds were maintained in an environmental temperature range of 26 ± 2 and $38\pm 2^\circ\text{C}$. Heterophil/lymphocyte ratio was investigated on day 1, 7, 14, 21 and 28 of the experimental period. The results revealed the following information: For those chickens maintained in an environmental temperature at $38\pm 2^\circ\text{C}$, the heterophil/lymphocyte ratio was higher than that of chickens at $26\pm 2^\circ\text{C}$. With the environmental temperature at $38\pm 2^\circ\text{C}$, the heterophil/lymphocyte ratio of the broilers was significantly higher than that of the Thai indigenous chicken crossbreds and Thai indigenous chickens ($p < 0.05$), respectively. The heterophil/lymphocyte ratio of the chickens for the environmental temperature of $38\pm 2^\circ\text{C}$ was significantly increased on day 7 and then significantly decreased to day 14 and 21 of experimental period ($p < 0.05$). This finding indicated that when chickens were maintained in high environmental temperatures, they were under heat stress. Chickens could adapt to high environmental temperatures. Finally, Thai indigenous chickens and Thai indigenous chicken crossbreds tolerated higher environmental temperatures than the broilers.

Key words: Heat stress, chronic, heterophil/lymphocyte ratio, Thai indigenous chickens, Thai indigenous chickens crossbred, broilers

INTRODUCTION

Stressors common to poultry production may include extreme environmental temperatures, disease, handling, beak trimming, vaccinations, crowding and inadequate ventilation (Altan *et al.*, 2000). Several studies have used heterophil/lymphocyte ratio as a measure of stress in birds (Al-Murrani *et al.*, 1997; Altan *et al.*, 2000; Carol *et al.*, 2000; Altan *et al.*, 2005). After birds are exposed to high ambient temperatures, the high heat creates workload on the physiological system (Aengwanich *et al.*, 2003a). When birds were under heat stress, the heterophil/lymphocyte ratio rose (Gross, 1989; Al-Murrani *et al.*, 1997; Altan *et al.*, 2000; Carol *et al.*, 2000). At present, the heterophil/lymphocyte ratio is generally accepted as the best indicator of stress in chickens (Altan *et al.*, 2000; Puvadolpirod and Thaxton, 2000) because it has been shown to be highly heritable and a reliable index (Gross and Siegel, 1983).

During the summer season in Thailand, environmental temperatures often reach $36-40^\circ\text{C}$ which is a dangerous temperature zone for broilers. Thai indigenous chickens, the wild birds that have been domesticated in the rural villages of Thailand over a long period of time, are familiar

with high environmental temperatures. Thai indigenous chickens, however, have a lower productive performance than broilers, so breeders have improved the production of the Thai indigenous chickens by crossbreeding them with chickens imported from overseas. Thai indigenous chicken crossbreds are a crossbred chicken of $\frac{1}{2}$ Thai indigenous chickens (cock), $\frac{1}{4}$ Rhode Island Red and $\frac{1}{4}$ Plymouth Rock (hen). Thai indigenous chicken crossbreds have a higher productive performance than Thai indigenous chickens. The purpose of this experiment was to compare tolerance to heat between Thai indigenous chickens, Thai indigenous chicken crossbreds and broilers by using the heterophil/lymphocyte ratio, a good marker for indication of heat stress in chickens. Results from this preliminary study would provide fundamental knowledge for improving poultry production by identifying a heat tolerant genetic resource for poultry production in tropical regions.

MATERIALS AND METHODS

Twenty four Thai indigenous chickens (12 males; 12 females), 24 Thai indigenous chickens crossbred (12 males; 12 females) and 24 broilers (12 males; 12 females),

one kilograms of weight and infectious disease-free were obtained from a commercial farm near Maharakham University and transferred to the laboratory of the Faculty of Technology, at Maharakham University. The experiment was performed during April-July, 2005. The experiments were begun after a 7-day adaptation period. The chicks were fed a standard ration *ad libitum* with continuous light and water supply. The experimental design was a split-split-plot design in CRD. The main plot involved two temperatures i.e., 26±2°C (continuous temperature) and 38±2°C (cyclic temperature; 26±2-38±2-26±2°C; chickens were maintained at 38±2°C for 6 h/day). The sub plot was 2x3 factorial i.e., sex (male and female) and 3 breeds of chicken (Thai indigenous chickens, Thai indigenous chickens crossbred and broilers). 6 Thai indigenous chickens, 6 Thai indigenous chickens crossbred and 6 broilers were maintained at each environmental temperature. On day 1, 7, 14, 21 and 28 of the experimental period, blood samples (via wing vein: 0.75 mL.) were collected and transferred to tubes containing EDTA as an anticoagulant (Ritchie *et al.*, 1994). Blood films were made, air dried and then stained with Giemsa-Wright's stain. The differential white blood cell (WBC) counts were performed by using the standard avian guideline of Ritchie *et al.* (1994). Heterophil/lymphocyte ratios were calculated (Gross, 1989). All data were analyzed by using the ANOVA procedure of Statistical Analysis System (1990). Means were separated by Duncan's multiple range tests. The level of significance was determined at p<0.05.

RESULTS AND DISCUSSION

Male and female Thai indigenous chickens, Thai indigenous chicken crossbreds and broilers were maintained at 26±2°C and 38±2°C. On day 1, 7, 14, 21 and 28 of experimental period, the heterophil/lymphocyte ratio of three breed chickens were determined. The results found that; on day 1, the heterophil/lymphocyte ratio of male and female Thai indigenous chickens, Thai indigenous chicken crossbreds and broilers maintained at 26±2°C and 38±2°C were not significantly different.

On day 7, the heterophil/lymphocyte ratio of broilers maintained at 38±2°C was significantly higher than that of Thai indigenous chickens and Thai indigenous chicken crossbreds at the same temperature and at the environmental temperature at 26±2°C (p<0.05). Moreover, the heterophil/lymphocyte ratio of female Thai indigenous chickens and Thai indigenous chicken crossbreds at 38±2°C was significantly higher than that at the environmental temperature at 26±2°C (p<0.05). The heterophil/lymphocyte ratio of the three breeds at 26±2°C was not significantly different.

On day 14, with the environmental temperature at 38±2°C, the heterophil/lymphocyte ratio of male broilers was significantly higher than that of female Thai indigenous chickens (p<0.05) and was significantly higher than that of Thai indigenous chickens, Thai indigenous chicken crossbreds and broilers maintained at 26±2°C (p<0.05). The heterophil/lymphocyte ratio of the three breeds at 26±2°C was not significantly different.

On day 21, the heterophil/lymphocyte ratio of male broilers maintained at 38±2°C was significantly higher than that of male and female Thai indigenous chickens and Thai indigenous chicken crossbreds (p<0.05). The heterophil/lymphocyte ratio of the three breeds at 26±2°C was not significantly different.

On day 28, the heterophil/lymphocyte ratio of the three breeds at 38±2°C was not significantly different (p>0.05). Furthermore, the heterophil/lymphocyte ratio of male broilers maintained at 38±2°C was significantly higher than that of male and female broilers at 26±2°C (p<0.05) (Table 1.)

The heterophil/lymphocyte ratio of the chickens at the environmental temperature of 38±2°C was significantly increased on day 7 and then significantly decreased on day 14 and 21 of experimental period (p<0.05). Moreover, the heterophil/lymphocyte ratio of chickens maintained at 38±2°C was significantly higher than that of chickens maintained at 26±2°C on day 7 and 14 (p<0.05) (Fig. 1.)

On day 28 of the experimental period, at 26±2°C, the heterophil/lymphocyte ratio of the Thai indigenous chicken crossbreds was significantly higher than the Thai indigenous chickens and broilers (p<0.05) and the

Table 1: Heterophil/lymphocyte Ratio (H/L ratio) of male and female Thai Indigenous Chickens (TIC), Thai Indigenous Chicken Crossbreds (TICC) and Broiler Chickens (BC) were maintained at 26±2°C and 38±2°C, on day 1, 7, 14, 21 and 28 of experimental period

| Parameters | Days | Environmental temperature at 26±2°C | | | | | | Environmental temperature at 38 ± 2°C | | | | | | SEM |
|------------|------|-------------------------------------|---------------------|--------------------|--------------------|--------------------|---------------------|---------------------------------------|--------------------|---------------------|---------------------|-------------------|---------------------|------|
| | | TIC | | TICC | | BC | | TIC | | TICC | | BC | | |
| | | Male | Female | Male | Female | Male | Female | Male | Female | Male | Female | Male | Female | |
| H/L ratio | 1 | 0.19 | 0.20 | 0.18 | 0.21 | 0.19 | 0.18 | 0.16 | 0.15 | 0.32 | 0.18 | 0.14 | 0.17 | 0.04 |
| | 7 | 0.23 ^d | 0.23 ^d | 0.29 ^{cd} | 0.24 ^d | 0.28 ^{cd} | 0.25 ^d | 0.34 ^{cd} | 0.55 ^b | 0.42 ^{bcd} | 0.47 ^{bc} | 0.82 ^a | 0.87 ^a | 0.06 |
| | 14 | 0.19 ^c | 0.24 ^{bc} | 0.20 ^c | 0.25 ^{bc} | 0.25 ^{bc} | 0.27 ^{bc} | 0.32 ^{abc} | 0.24 ^{bc} | 0.29 ^{abc} | 0.33 ^{abc} | 0.42 ^a | 0.36 ^{ab} | 0.04 |
| | 21 | 0.21 ^{bc} | 0.17 ^{abc} | 0.26 ^{bc} | 0.24 ^{bc} | 0.24 ^{bc} | 0.29 ^{abc} | 0.21 ^{bc} | 0.19 ^c | 0.23 ^{bc} | 0.24 ^{bc} | 0.37 ^a | 0.31 ^{ab} | 0.03 |
| | 28 | 0.25 ^{abc} | 0.25 ^{ab} | 0.41 ^{ab} | 0.35 ^{ab} | 0.15 ^{bc} | 0.06 ^c | 0.32 ^{ab} | 0.33 ^{ab} | 0.37 ^{ab} | 0.35 ^{ab} | 0.50 ^a | 0.27 ^{abc} | 0.08 |

^{a, b, c and d} within row, mean with no common superscript(s) differ significantly (p<0.05), SEM = Standard error of the mean

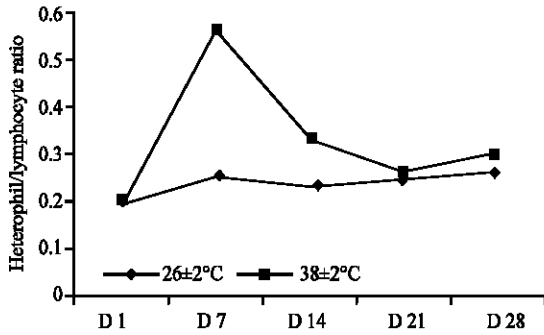


Fig. 1: Heterophil/lymphocyte ratio pattern of chickens maintained in the environmental temperature at 26±2°C and 38±2°C on days 1, 7, 14, 21 and 28 of experimental period

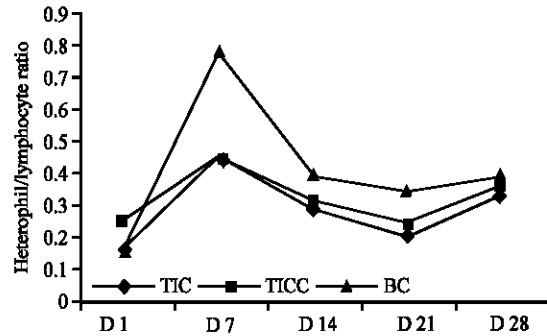


Fig. 3: The heterophil/lymphocyte ratio pattern of chickens Thai Indigenous Chicken (TIC), Thai Indigenous Chicken Crossbreds (TICC) and broilers (BC) on days 1, 7, 14, 21 and 28 of experimental period at 38±2°C

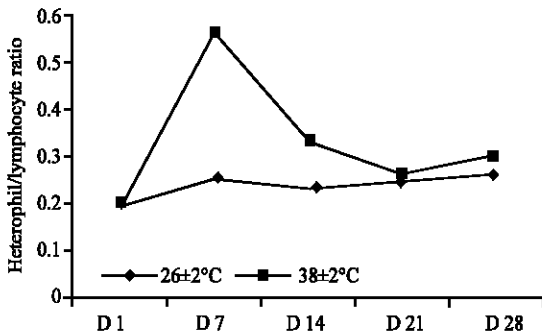


Fig. 2: The heterophil/lymphocyte ratio pattern of chickens (Thai indigenous chicken (TIC), Thai indigenous chicken crossbreds (TICC) and broiler (BC)) on days 1, 7, 14, 21 and 28 of experimental period at 26±2°C

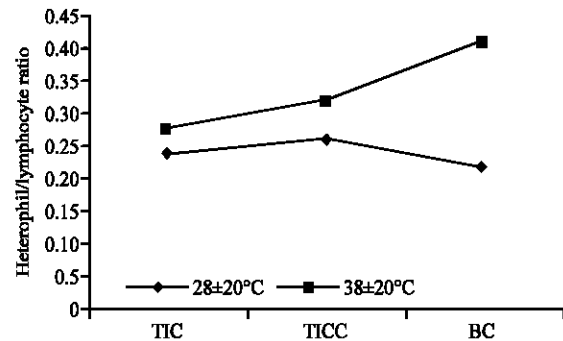


Fig. 4: Heterophil/lymphocyte ratio pattern of chickens (Thai indigenous chicken (TIC), Thai indigenous chicken crossbreds (TICC) and broilers (BC)) with the environmental temperature at 26±2°C and 38±2°C

heterophil/lymphocyte ratio of Thai indigenous chickens was significantly higher than the broilers ($p < 0.05$) (Fig. 2.)

On day 7 of the experimental period, the heterophil/lymphocyte ratio of the broilers maintained in the environmental temperature at 38±2°C was significantly higher than that of the Thai indigenous chickens and the Thai indigenous chicken crossbreds ($p < 0.05$). In addition, the heterophil/lymphocyte ratio significantly increased on day 7 and then significantly decreased to day 14 of experimental period ($p < 0.05$) (Fig. 3.)

With the environmental temperature at 38±2°C, the heterophil/lymphocyte ratio of the broilers was significantly higher than that of the Thai indigenous chicken crossbreds and the Thai indigenous chickens ($p < 0.05$) (Figure 4.)

When chickens were maintained in an environmental temperature at 38±2°C, their heterophil/lymphocyte ratio was higher than that of chickens at 26±2°C and they were under heat stress. This result is in accord with reports of

Gross (1989), Aengwanich (2002), Aengwanich and Chinrasri (2002), Aengwanich and Simaraks (2002), Aengwanich and Simaraks (2003), Aengwanich *et al.* (2003a) and Aengwanich *et al.* (2003b). They found that the heterophil/lymphocyte ratio of chickens maintained in high environmental temperatures increased. Jain (1993) explained that glucocorticoid, which is released in large amounts when chickens are under stress, causes heterophilia primarily by inducing the increased release of heterophils from the bone marrow through the circulation. On the other hand, Jain (1993) also explained that glucocorticoid induced lymphopenia is attributed to, lympholysis in blood, DNA damage, lymphoid tissue atrophy and increased shift of lymphocytes from the blood to other body compartments. These explanations show that the heterophil/lymphocyte ratio in blood circulation increases.

With the environmental temperature at $38\pm 2^{\circ}\text{C}$, on day 1, the heterophil/lymphocyte ratio of the Thai indigenous chickens, Thai indigenous chicken crossbreds and broilers was not significantly different, because samples were collected before they were exposed to high heat. On day 7, 14 and 21, the heterophil/lymphocyte ratio of the broilers was significantly higher than the Thai indigenous chickens and the Thai indigenous chicken crossbreds. Whereas, on day 28, the heterophil/lymphocyte ratio of the three breeds of chickens significantly decreased to normal range and the heterophil/lymphocyte ratio of the broiler, the Thai indigenous chicken and the Thai indigenous chicken crossbreds were not significantly different. These results indicated that the broilers were less tolerant to high heat than the Thai indigenous chickens and Thai indigenous chicken crossbreds. Moreover, the heterophil/lymphocyte ratio of the Thai indigenous chickens and Thai indigenous chicken crossbreds was not significantly different. This phenomenon showed that the crossbred chicken did not have a significantly different tolerance to high heat than the Thai indigenous chicken. The heterophil/lymphocyte ratio of chickens of both sexes in high environmental temperature conditions was not significantly different, but on day 7, the heterophil/lymphocyte ratio of the female Thai indigenous chickens was significantly higher than that of the male. This occurrence only showed that during early exposure to high heat, the female Thai indigenous chicken was less tolerant to the high heat than the male. On day 17, 21 and 28 of the experimental period, the heterophil/lymphocyte ratio of the male and female Thai indigenous chickens were not significantly different.

At $38\pm 2^{\circ}\text{C}$, the heterophil/lymphocyte ratio of chickens was significantly increased on day 7 and then significantly decreased on day 14 and then was not significantly different on day 21 of experimental period. Moreover, on day 28, the heterophil/lymphocyte ratio of chickens at $26\pm 2^{\circ}\text{C}$ was not significantly different from chickens maintained at $38\pm 2^{\circ}\text{C}$. These results are similar with the report of Puvadolpirod and Thaxton (2000); they found that chickens could adapt to stress conditions. Therefore, results from this experiment showed that when chickens were under heat stress, they could adapt to high heat and Thai indigenous chickens and Thai indigenous chicken crossbreds were more tolerant to high environmental temperatures than broilers.

CONCLUSIONS

Thai indigenous chickens, Thai indigenous chicken crossbreds and broilers were maintained at the

environmental temperature at $26\pm 2^{\circ}\text{C}$ and $38\pm 2^{\circ}\text{C}$ and then a heterophil/lymphocyte ratio was determined for comparison of the chicken's ability to tolerate heat. The results found that when chickens were maintained in an environmental temperature at $38\pm 2^{\circ}\text{C}$, their heterophil/lymphocyte ratio was higher than that of chickens at $26\pm 2^{\circ}\text{C}$ and they were under heat stress. Chickens could adapt to high environmental temperatures. In addition, Thai indigenous chickens and Thai indigenous chicken crossbreds had a greater tolerance to high environmental temperatures than broilers.

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