HEAT STRESS INDUCES HISTOPATHOLOGICAL CHANGES IN LYMPHOID ORGANS OF BROILER AND PHILIPPINE NATIVE CHICKENS

Ma. Suzanneth G. Lola¹, Michelle Grace V. Paraso², Billy P. Divina¹, Delia A. Gobonseng¹, Ariel M. Bombio² and Therese Marie A. Collantes¹

ABSTRACT

This study was conducted to describe histopathologic changes in lymphoid organs of broiler and native chickens in response to heat stress. A total of 60 day-old broiler and native chicks were allocated in 3 treatment groups. The control group (T1) was exposed to ambient environmental temperature (AET); Treatment 2 (T2) was exposed to AET + ≈1°C; and T3 was exposed to AET + ≈3°C. Each treatment was exposed to the required temperature for 7 hours from d 29 to 35 (AET = 29.8°C) and from d 43 to 49 (AET = 31.7°C). Results showed significant microscopic changes in the lymphoid organs that include lymphoid depletion in the Bursa of Fabricius and lymphoid depletion and lymphocytic degeneration in the germinal centers of the spleen. The thymus had minimal to mild degenerative changes mainly in the cell population of the medulla. The results also showed more pronounced microscopic changes in the Bursa of Fabricius and spleen of broiler chicks compared to native chicks. The degree of response to heat stress appears to be breed-dependent with the lymphoid organs of broiler chickens showing lesser adaptive capacity to withstand high temperature than the lymphoid organs of native chickens.

Keywords: broiler, heat stress, histopathology, lymphoid organs, Philippine native chicken

INTRODUCTION

The poultry industry is considered as the fastest growing sector on meat production worldwide. In the Philippines, the demand for poultry products is generally optimistic but always challenged by threats from less efficient production systems and higher input costs (Chang, 2004). A key driver in the latter is maintenance of flock health, which encompasses prevention, treatment and control of disease. Disease occurrence translates to adverse impacts on farm growth and profitability.

One of the major challenges that currently confront poultry producers is the extreme temperature brought about by climate change. The detrimental effects of high ambient temperature on the efficiency of broiler production and meat yield has been established (Sandercock et al., 2001); whereas in laying hens, it has been known to reduce egg production and quality (Lara and Rostagno, 2013; Mashaly et al., 2004). These effects have been attributed to the adverse effects of heat stress on the metabolism and behavior of the animal, leading to lower feed intake and productivity. The economic problems associated with heat stress could be devastating particularly to smallholders who have no access to temperature-controlled housing facilities.

Aside from the economic impacts of stress on animal production, much of the research on the adverse consequences of heat stress on animal production has been conducted due to concerns on animal welfare (Mashaly et al., 2004; Quinteiro-Filho et al., 2010). The severity of response of the animal to heat stress may vary depending on
its breed, species, physiological and nutritional status, and genetic potential (Devendra, 2012). Although chickens undergo thermoregulatory adaptations during periods of heat stress (Lara and Rostagno, 2013), studies showed that exposure to high ambient temperatures either in intensity (acute heat stress) or over long periods (chronic heat stress) could suppress the immune system and may lead to death if coupled with high humidity (Butcher and Miles, 2015). High mortality rates have been observed in local intensive poultry farming systems as a result of high ambient temperature and relative humidity (Lambio, 2010).

A strategy used by some poultry producers to cope with the negative economic impacts of climate change is to raise climate tolerant breeds. Although slower in terms of growth, native chicken breeds in tropical countries are generally recognized as more resilient to heat stress (Soeleiman et al., 2011). Exotic and fast-growing strains generate more body heat as a result of faster metabolism but somehow fail to achieve effective heat dissipation under hot and humid environmental conditions (Cahaner, 2008). However, there are gaps in knowledge on the underlying mechanisms behind the perceived resilience of indigenous local chicken breeds under hot environmental conditions. This study was conducted to compare the histopathological changes in the following lymphoid organs of broiler and Philippine native chickens in response to heat stress: thymus, spleen and Bursa of Fabricius, which are vital components of the avian immune system.

**MATERIALS AND METHODS**

**Management of birds**

Procedures in this study were approved by the Institutional Animal Care and Use Committee of the College of Veterinary Medicine, University of the Philippines Los Baños (UPLB). A total of 60 day-old broiler chicks (White leghorn) and native day-old chicks were used in the study. The broiler chicks (n=30) were obtained from a commercial hatchery whereas the native day-old chicks (n=30) of the Banaba (BNB) (n=15) and Paraokan (PRK) (n=15) breeds were purchased from the National Swine and Poultry Research and Development Center (NSPRDC) of the Bureau of Animal Industry (BAI) Animal Station in Tiaong, Quezon. The chicks were kept in the experimental animal house of the UPLB Veterinary Teaching Hospital – Maahas Station, reared in raised one-tier battery cages with wire flooring and were subjected to standard husbandry and management procedures of growing broiler chickens. During the weeklong brooding period, the birds were maintained on a booster diet. From d 8 to d 28, the birds were provided access to starter mash and thereafter, to grower ration for the remainder of the 86 87

**Heat stress induction**

From d 1 to d 27 of age, the birds were maintained under ambient environmental temperature (AET). On d 28, each breed was randomly allocated into 3 different treatments that were separated from each other by nylon canvass walls. The chickens were then randomly distributed into groups of 5 for the broilers and groups of 10 for the native chickens. The control group (T1) was exposed to ambient environmental temperature (AET); Treatment 2 (T2) was exposed to AET + ≈1°C; and T3 was exposed to AET + ≈3°C. The increase in temperature in T2 and T3 was accomplished with the use of incandescent light bulbs and electric heaters. Chickens were exposed to the desired temperature for 7 hours from d 29 to d 35 (AET = 29.8°C) and from d 43 to d 49 (AET = 31.7°C). The temperature and relative humidity in each treatment were monitored and recorded in all areas using a digital thermohygrometer. The relative humidity was not controlled and ranged from 65-70 (T1), 64-68 (T2) and 48-65 (T3) on d 29 to d 35; and from 54-58 (T1), 54-56 (T2) and 49-52 (T3) on d 43 to d 49.

**RESULTS AND DISCUSSION**

The effects of heat stress on the microscopic features of the thymus, Bursa of Fabricius and spleen are shown in Figure 1. Major histopathological changes were observed in the Bursa of Fabricius of both native and broiler chickens exposed to the highest temperature (T3). Majority of the broiler chickens showed atrophy of the Bursa of Fabricius characterized by depletion of lymphoid cells in medulla, intrafollicular edema and decreased cortical width. Among native chickens, depletion of lymphoid cells in the medulla was the only lesion observed. These histopathological changes were consistent with the findings of Anju Rajan et al. (2014). Aengwanich (2008), on the other hand, obtained a low number of lymphocytes not only in the medulla but also in the cortex of the bursa of native chickens. For both broiler and native chickens in T1 and T2, the Bursa of Fabricius showed no distinct histopathological changes.

The spleens of both native and broiler chickens in T3 likewise presented major morphological changes. Most of the spleens in both breeds showed mild to moderate lymphoid cell depletion in the germinal centers of the white pulp. In T2, some broiler chickens had spleens with moderately depleted lymphoid cells in the germinal centers of the white pulp. In comparison, the spleens of native chickens in T2, and both broiler and native chickens in T1 presented no distinct histopathological changes. In another study, normal histology of spleen was observed even after exposure to high temperatures (Anju Rajan et al. 2014).

Thymi from both broiler and native chickens in T1 and T2 generally showed no distinct histopathological changes. In contrast, some thymi of native and broiler chickens in T3 presented mild degenerative changes in the cell population of the thymus with increased eosinophilic colloid materials. These degenerative changes in the thymus were not consistent with the result obtained on heat-stressed chickens by Anju Rajan et al. (2014).

The thymus and Bursa of Fabricius are considered as primary lymphoid organs that serve as sites for lymphocyte generation and growth. The spleen, on the other hand, is a secondary lymphoid organ where mature and functional T- and B- lymphocytes accumulate and come into contact with specific antigens (Grogan et al., 2008). Based on our results, these organs demonstrated susceptibility to the effects of heat stress with the broiler spleen developing lesions even at ambient temperature levels. In native chickens, the thymus was the lymphoid organ to initially manifest lesions at T2 temperature. Lesions were observed to be present in all lymphoid organs after exposure to T3 temperature. Furthermore, the lesions seen in broiler chickens were more pronounced compared to the changes seen in native chickens.

In vertebrates, the hypothalamic-sympathetic and the hypothalamic-pituitary-adrenal (HPA) systems provide brain and peripheral control of stress responses. Stimulation
Heat stress effects on lymph nodes of Philippine native and broiler chickens

Figure 1. Bursa of Fabricius (A-B), spleen (C-D) and thymus (E-F) of broiler (BC) and/ or Philippine native chickens (NC) subjected to ambient environmental temperature (AET) (T1), AET + ≈1°C (T2) and AET + ≈3°C (T3). (A) Bursa of Fabricius in BC showing no distinct histopathological changes, T1; (B) Moderate depletion of lymphoid cells in the medulla, intrafollicular edema and decreased cortical width in the bursa of Fabricius of BC, T3; (C) Spleen in NC showing no distinct histopathological changes, T1; (D) Moderate depletion of lymphoid cells in the germinal center of the splenic white pulp NC, T3; (E) Thymus of NC showing no distinct histopathological changes, T1; (F) Mild degenerative changes in the medullary cells and increased eosinophilic colloid materials in the thymus of NC, T3. H&E stain. (marker of A to B = 200 µm; marker of C to F = 50 µm).

of the HPA axis due to stressful stimuli causes the release of glucocorticoids, which have been demonstrated to affect the immune system (Salak-Johnson and McGlone, 2007). Heat stress has been found to decrease the size and weight of lymphoid organs in broiler chickens as documented in previous studies (Bartlett and Smith, 2003; Niu et al., 2009 and Quinteiro-Filho et al., 2010). Tarek et al. (2013) had the same observation in the bursa of Fabricius of heat-stressed broilers in addition to histopathological changes that were reported in this study.

Involution is the most frequently encountered change that can be induced by heat stress on lymphoid organs (Puvadolpirod and Thaxton, 2000). High serum cortisol levels have been associated with apoptotic effect on lymphoid cells (Sandhu et al., 2012) eventually leading to tissue depletion and reduction of the avian resistance to disease. In this study, the observed histopathological changes in the examined organs, i.e., lymphoid cell depletion especially in T2 and T3 are indicative of involution. On the other hand, the increased eosinophilic colloids in the thymus of chickens in T3 are suggestive of high secretions in this organ. Thymic secretions have increased serum corticosterone levels in rodents (Cooper 1984; McGillis et al., 1985) and could have influenced glucocorticoid production in response to heat stress in this study.

In conclusion, this study demonstrated that exposure to temperatures approximately 30°C and above are sufficient to induce pathological lesions in the lymphoid organs of both native and broiler chickens. However, the degree of response to heat stress appeared to be breed-dependent with the lymphoid organs of broiler chickens showing lesser adaptive capacity to withstand higher ambient temperatures than the lymphoid organs of native chickens. The gene coding for adaptive capacity to environmental stressors may have been better conserved in this breed, thus enabling these birds to withstand the adverse health effects of constant exposure to heat stress.

ACKNOWLEDGMENT

The authors would like to thank the Philippine Commission on Higher Education through its DAP GIA RDE Program for funding this study.

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