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EFFECT OF SEASON AND DIETARY PROTEIN LEVEL ON IMMUNE RESPONSE OF THREE EXOTIC BROILER STRAINS IN SUDAN

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ABSTRACT: *This study was conducted to investigate the effect of the season (summer versus winter) and dietary protein level (high versus low) using three broiler strains (Ross, Cobb and Hubbard) on immunity; heterophil/lymphocyte ratio and haemagglutination against sheep red blood cells (SRBC). Three hundred and sixty, one-day-old unsexed broiler chicks were used in this study during the summer and winter seasons, 120 from each of Ross strain, Cobb strain and Hubbard strain. Two experiments were executed in a complete randomize design (factorial arrangement 3x2x2). Each strain was divided into two groups, with six replicates (10 chicks per replicate). Group A of each strain was fed on a starter diet containing 23% crude protein for the first four weeks of age, then replaced by a finisher diet containing 21% crude protein. Group B was fed on a starter diet containing 21% crude protein replaced by a finisher diet containing 19% crude protein. Both diet were iso caloric. The results showed that the heterophil/lymphocyte ratio (H/L ratio) increased significantly (P<0.05) during the summer in both Hubbard and Ross strains, but it was not significantly affected by the season in Cobb strain. The total antibody titers against SRBC were decreased during the summer season in the three strains. The level of dietary protein showed no significant effect on H/L ratio in the three strains. Decreasing dietary protein level decreased the total antibody titers against SRBC in both Ross and Hubbard strains. Whereas, it does not affect the total antibody titers against SRBC in Cobb strain.*

ORIGINAL ARTICLE

Key words: *Broiler Strain; Season; Protein Level; Immune Response*

INTRODUCTION

Over the last several decades, genetic selection for faster growth rate, better feed efficiency and higher disease resistance are intensively considered in commercial broiler production. Measures of immunity that have been commonly used and assessed in poultry are antibody response to foreign antigens (Patterson and Siegel, 1998).

The phenotype of an individual represents the complex sum of the effects of genotype and environment. Many studies on interactions between genetic factors and environments (G × E) have been reported for chickens (Ali et al., 2001; Deeb and Cahaner, 2001a, b, 2002; Tixier-Boichard, 2002; Mathur, 2003; Fulton, 2004).

Despite progress that has been made in the areas of health, vaccination and management in the poultry industry, diseases problem still exist that affect the efficiency of the poultry industry. Seasonal cycles of infectious diseases have been variously attributed to changes in environmental conditions, the prevalence or virulence of the pathogen, or the behaviour of the host, but no single theory has proved a satisfactory explanation (Dowell 2001).

Several studies have been conducted on the effects of high temperature on the immune responses of chickens, with variable findings. Thaxton et al. (1968) was the first who demonstrated that high environmental temperature affects the development of specific immune responses in young chickens.

The effect of nutrition on antibody response to Sheep Red Blood Cells (SRBCs) is variable. Tsiagbe et al. (1987) reported a dose related increase in total and immunoglobulin-G (IgG) antibodies against SRBCs when the broiler chicken diet was supplemented with methionine. However, Rao et al. (1999) found no significant differences in humoral response to SRBCs among the chicks fed high, medium and low protein diets. Similarly, dietary protein and energy content have no significant influence on broiler chick responses to SRBCs (Praharaj et al., 1997). Nevertheless, Carlomagno et al. (1980) reported that protein deficiency inhibited antibody production and the development of antibody production cells in response to T-dependent antigens.

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MATERIAL AND METHODS

Two experiments were carried out in the premise of poultry research unit, department of poultry production, Faculty of Animal Production University of Khartoum, Shambat (Khartoum North). The laboratory analyses were carried out at the Department of Physiology, Faculty of Veterinary Medicine and University of Khartoum.

Experimental birds

Three hundred and sixty one-day-old unsexed broiler chicks were used during the summer and the winter seasons, 120 from Ross, 120 for Cobb and 120 for Hubbard strain. The total number of chicks of each strain was divided into two groups, with six replicates (10 chicks per each).

Experimental diets

Group A of each strain was fed a diet containing 23% crude protein and 3000 kcal/kg ME as starter diet for the first four weeks of age, and then replaced by a diet containing 21% crude protein and 3000 kcal/ kg ME as finisher diet. Group B for each strain was fed a diet containing 21% crude protein and 3000 kcal/ kg ME as starter diet for the first four weeks and then shifted to a diet containing 19% crude protein and 3000 kcal/ kg ME as finisher diet. The formulation of the experimental diets is shown in table 1.

Table 1 - Ingredients composition of experimental diet on percent basis

Ingredient	Diet (1)		Diet(2)	
	Starter %	Finisher %	Starter %	Finisher %
Sorghum	61	61.5	61.5	68.5
Groundnut cake	15.8	12	12	7.3
Sesame cake	13	11.3	11.3	9
Wheat bran	4	9	9	9
Super concentrate	5	5	5	5
Limestone	0.9	0.9	0.9	0.9
Nacl	0.25	0.25	0.25	0.25
Lysine	0.04	0.04	0.04	0.04
Methionine	0.01	0.01	0.01	0.01
Total	100	100	100	100

* Composition of supper concentrates BRO-5 (1504.10) Fishmeal, vegetable protein, dicalciumphosphate, limestone, vitamins, trace-elements, antioxidant. * Vitamins and premix minerals per kg of diet

Heterophils / Lymphocyte ratio

Heterophils / Lymphocytes (H/L) ratio was calculated using blood smears after been stained by Giemsa May-Grunwald staining procedure.

Antibody response to SRBC

The Sheep Red Blood Cells (SRBC) were collected and washed three times in normal saline. Birds were injected intravenously with 1 ml of 10 % suspension of packed sheep red blood cells in normal saline at four weeks of age. 11 days later, blood was collected from the heart of each bird. Sera were collected and stored at -20°C. The total antibody titre was determined by haemagglutination test. Briefly, 50 µL of plasma was added in an equal amount of phosphate buffer solution in the first column of a 96-well U-shaped bottom plate and the solution was incubated for 30 min at 37°C. A serial dilution was then made and 50 µL of 2% SRBC suspension was added to each well. Total antibody titers were then read. The reciprocal of the highest dilution showing complete agglutination was expressed as titre (log2).

Statistical Analysis

Haemagglutination antibody titers against SRBC were statistically converted into geometric mean titers. The statistical analysis for the recorded H/L ratio was carried out using analysis of variance for factorial experiment in a completely randomized design by general linear model using (Statistix program, version 9).

RESULTS AND DISCUSSION

The findings of the present study indicated that there was a significant ($P<0.05$) increase in H/L ratio in Ross and Hubbard strains during summer compared to winter season. While, Cobb strain was not significantly affected by heat stress during summer season (Figure1). The stability of H/L ratio in Cobb strain irrespective of the season could be attributed to the high immune response of Cobb compared to the other two strains (Makram et al*.,* 2010). The increased in H/L ratio during the summer might be due to induced stress which release glucocorticoids, causing dissolution of lymphocytes in lymphoid tissues, leading to lymphopenia. However, there is an increase in heterophil released by the bone marrow, thus increasing their number in the circulation, although their phagocytic and bactericidal activities are decreased (Swenson and Reece, 1996; Berne and Levy, 1998). The present finding is in consistent with the previously obtained ones (Zulkifli and Siegel, 1995; Borges, 1997; Altan et al*.,* 2000; Bedenova et al., 2003; Zulkifli et al*.,* 2009).

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The data concerning the effect of broiler strain and season on the antibody titers against SRBC are presented in (Figure 2). The results showed that the heat stress caused a reduction in total antibody titers against SRBC in all strains. However, it was less in Cobb strain. This result is in accordance with the findings of Zulkifi et al. (2000) who found that heat stress caused a reduction in antibody synthesis. This reduction could be indirectly due to an increase in inflammatory cytokines under stress (Ogle et al., 1997), which stimulates the hypothalamic production of corticotropin releasing factor (Sapolsky et al., 1987). Corticotropin releasing factor is known to increase adrenocorticotropic hormone from the pituitary; adrenocorticotropic hormone then stimulates corticosterone production from the adrenal gland. Corticosterone inhibits antibody production (Gross, 1992). Furthermore, heat stress is known to decrease T-helper 2 cytokines (Wang et al., 2001), which are important for antibody production, (Lebman and Coffman, 1988).

The results reflected that, in all strains the level of dietary protein had no significant effect on H/L ratio (Figure 3). This result is in the line with that obtained by Donkoh et al. (1999); Alam et al. (2004) who observed that the level of dietary protein has no significant effect on H/L ratio. Furthermore, feeding birds with diets containing high dietary protein increased the total antibody titers against SRBC. This result is in consistent with those of (Payne et al., 1990) who found that deficiency or excess of dietary protein changes immune responses. Deif et al. (2007) documented that the total anti-SRBCs antibody titers measured post primary and secondary SRBCs-injection for broiler chicks fed a high protein diet are significantly higher than those of other fed a marginal protein diet which is in agreement with the findings of this study.

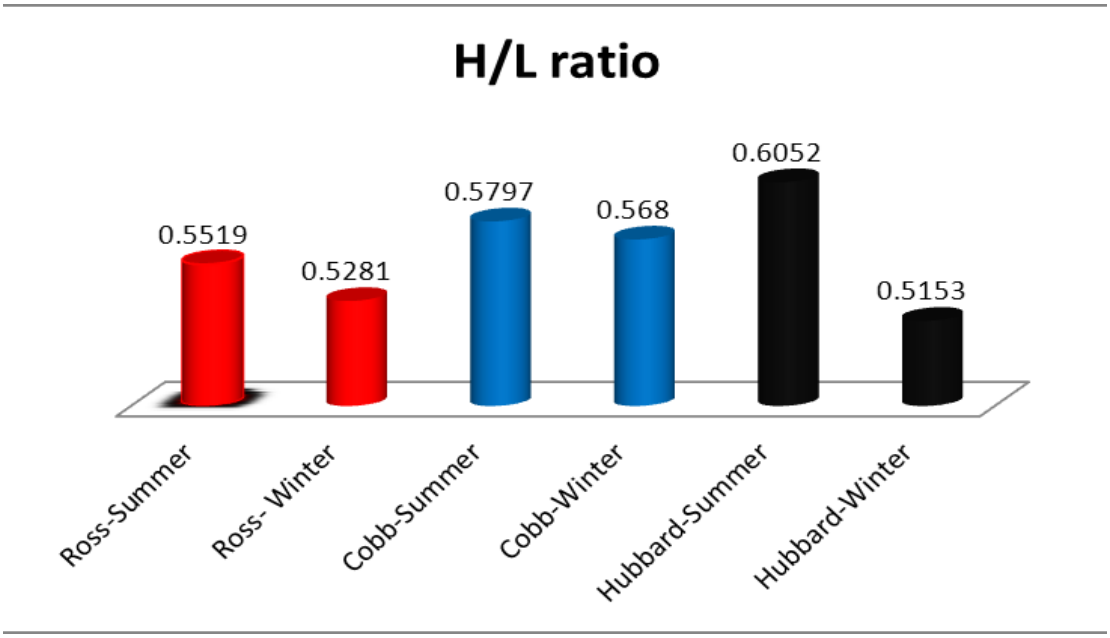

Figure 1 - Effect of broiler strain and season on Heterophils/ Lymphocyte ratio

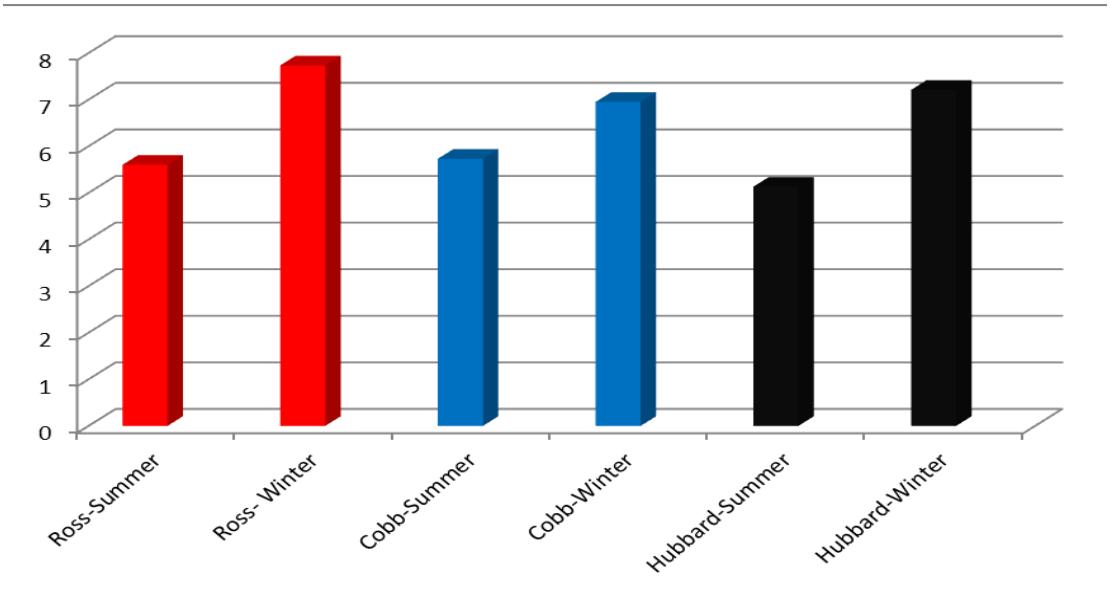

Figure 2 - Effect of broiler strain and season on total antibody titers against SRBC

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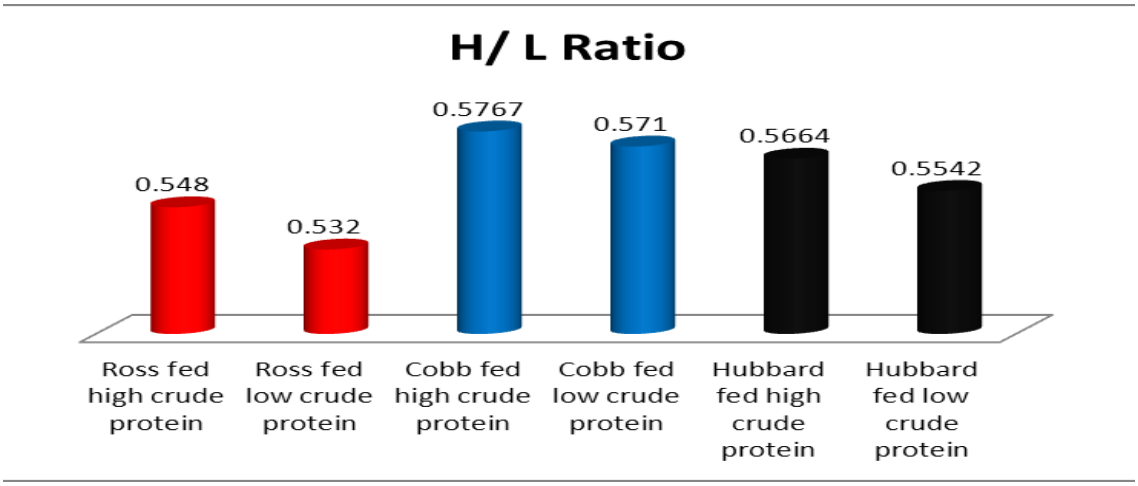

Figure 3 - Effect of broiler strain and dietary protein level on H/L ratio

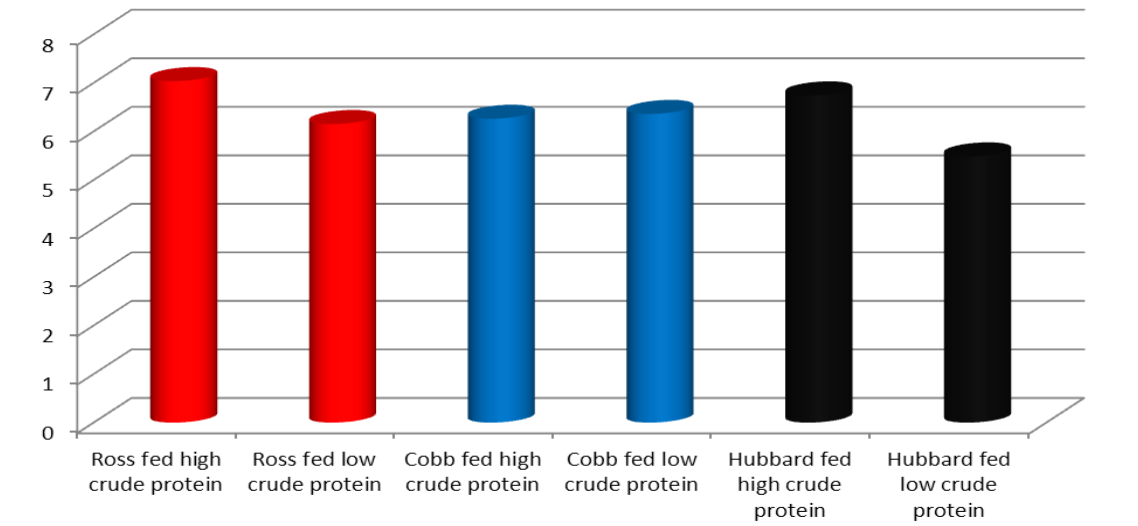

Figure 4 - Effect of broiler strain and dietary protein level on total antibody titers against SRBC

CONCLUSIONS

The physiological responses of genetically improved broiler strains to environment (heat stress) and nutritional (low protein) stresses appeared to be significantly different from each other. Cobb strain seemed to be more tolerance to summer heat stress in tropical areas such as in Sudan (under open-sides system).

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