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Stress Biomarkers in Vanaraja Chicken Maintained Under Various Rearing Systems

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Abstract

Stress is of major concern for poultry industry because it exerts deleterious effects on different parameters like feed intake, feed conversion ratio, weight gain, etc. In present study various enzymatic viz. superoxide dismutase (SOD), catalase (CAT), glutathione peroxidase (GSH-Px) and non-enzymatic components like reduced glutathione (GSH), hemoglobin and stress induced cellular damage i.e. lipid peroxidation was estimated to access the stress level in Vanaraja chickens reared under various rearing systems during summer. Significantly ($p<0.05$) increased activities of CAT and SOD was observed in deep litter system as compared to cage and semi-intensive rearing system. However, non-significant change in CAT and significantly increased activity of SOD was observed as the age progress. GSH-Px activity significantly lower ($p<0.05$) in the deep litter as compared to other systems, however, the activity increases significantly ($p<0.05$) at 8th wks as compared to 4th wks. GSH level was found maximum in cage system compared to deep litter and semi-intensive system. Non-significant changes were observed in hemoglobin concentration during study both between age groups as well as the age progresses. Observations of the study suggested that cage system is better than deep litter and semi-intensive system in handling the stress induced by different environmental factors.

Keywords: Chicken; biomarkers; chicken

Introduction

Poultry industry plays a major role in food security for the human population and its production can be maximize by providing them comfortable environment to birds. Due to their short production cycle and high feed efficiency, poultry breeds are more susceptible to changing environmental conditions and diseases than other domestic species (Nolan *et al*., 1999, Fatufe *et al*., 2004). Change in environmental temperature, relative humidity, light, transport, environmental pollutants, ventilation and rearing system significantly affect the growth and production of poultry (Campbell and Lasley, 1975, Betteidge, 2002). Out of these, type of rearing system also play major role for the faster growth, better feed conversion ratio, least mortality etc., thus providing maximum benefit to the farmers. In India, mainly three rearing systems have been adopted i.e. cage, deep litter and semi-intensive depend on breeds characteristics.

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Vanaraja a dual purpose strain, required low metabolizing energy, better productivity, sustainability and adaptability in rural areas was developed by Project Directorate on Poultry (PDP) Hyderabad, India (Rama Rao *et al*., 2005, 2007). Stress reduces feed intake, body weight gain (Geraert *et al*., 1996), carcass yield (McGeehin and Mirabelli, 2001), egg production (Smith, 1993), immunosuppression and increase disease susceptibility to birds (Yahav *et al*., 1995). To maximize production, Vanaraja chicken should be kept in least stress condition especially in the hot and humid regions of the world. Thus, the present study was aimed to determine the levels of stress biomarkers in Vanaraja chicken reared in different rearing systems like semi-intensive (deep liter and cage system) and intensive system of rearing with the changing weather conditions.

Materials and methods

Experimental protocol

Day old chicks of Vanaraja were procured from the Department of Animal Husbandry, Jammu and

Kashmir. The chicks were followed a uniform pattern of brooding, vaccination, feeding and watering of all the birds up-to two wks of age. Strict hygienic and sanitation procedures were adopted during the study. After two wks of brooding, chicks (120) were randomly divided into three groups of 40 chicks with 4 replicates of 10 chicks each. Group I reared in cage system, group II kept in the deep litter system whereas, group III chicks were reared in semi-intensive system. The chicks were fed with normal diet Crumbre starter supplied by the Godrej Agrovet Ltd. India. The study was conducted during summer (May & June, 2008) and during study period environmental temperature and humidity was recorded by University Metrological Centre. Temperature humidity index (THI) was calculated as per the method described by the Mc Dowell (1972).

Stress biomarkers assays

Blood was collected directly from the heart using 18 gauge needles in clean sterile heparinised tubes from 2 chicks of each replicates and a total of 8 chicks from each group, after 2 weeks of rearing in different system (4th week of age). However, at 8th week blood was collected from the tarsal vein. Whole blood is used for the estimation of blood glutathione (GSH) and hemoglobin. Erythrocyte lysate (1%) was used for the CAT, SOD, GSH-Px whereas, 33% lysate was used for the determination of lipid peroxidation activity of the erythrocyte membrane. The extent of lipid peroxidation was estimated as the concentration of thiobarbituric acid reactive products (malondialdehyde) by the method of Ohkawa *et al*. (1979). The activity of SOD and CAT was measured according to the method described by Marklund & Marklund (1974) and Aebi (1983) respectively. The GSH-Px and GSH activity in erythrocyte lysate was assayed by the methods of Hafeman *et al*. (1974) and Beutler(1975) respectively.

Statistical analysis

The data were expressed as mean ± SE. Statistical analyses were done by one-way ANOVA followed by Dunnet's test with $P \leq 0.05$ as a limit of significance.

Results

In present study various enzymatic components viz. SOD, CAT, GSH-Px and non-enzymatic components like GSH, hemoglobin are estimated to access the stress level in Vanaraja chickens reared in different systems were presented in the table 1. The change in the THI twice in a day (morning and evening) during study period was estimated and their graphical representation was shown in fig. 1. Average THI values in morning and evening in first

Table 1. Alteration in stress biomarkers and lipid peroxidation in different rearing system of Vanaraja chicken

Stress Parameters (Units)	Age group of chicks					
	4th weeks			8th weeks		
	Group I	Group II	Group III	Group I	Group II	Group III
Catalase (µmole of H2O2 decomposed min-1 mgHb-1)	32.37±2.56a	40.97 ±3.65b	22.23±2.03a	25.82±2.52a	24.08±4.34a	34.67±3.74a
SOD (Units mgHb-1)	0.057±0.005aB	0.07±0.004aA	0.024±0.010aB	0.17±0.007b	0.175±0.005b	0.169±0.008b
GSH-Px (Units mgHb-1)	6.20±0.21aB	4.51±0.48aA	6.97±0.11aB	10.84±0.27b	10.92±0.42b	10.66±0.26b
GSH (n moles ml-1)	106.01±9.36C	37.64±3.29aA	73.991±8.39aB	108.26±10.59A	101.85±1.79bA	125.18±10.56bB
LPO (n moles MDA produced gm Hb-1 hr-1)	4.49±0.64A	2.89±0.26aB	2.475±0.14aB	5.31±0.29B	3.81±0.59bA	4.84±0.98bB
Hb (gm %)	9.43±0.38a	10.78±0.37a	11.15±0.35a	10.92±0.37a	10.72±0.35a	10.59±0.41a

Values are expressed as mean ± S.E. of 8 birds

Cap. Alphabets: different superscript differs significantly between groups within a time period.

Small Alphabets: different superscript differs significantly between time periods with in a group.

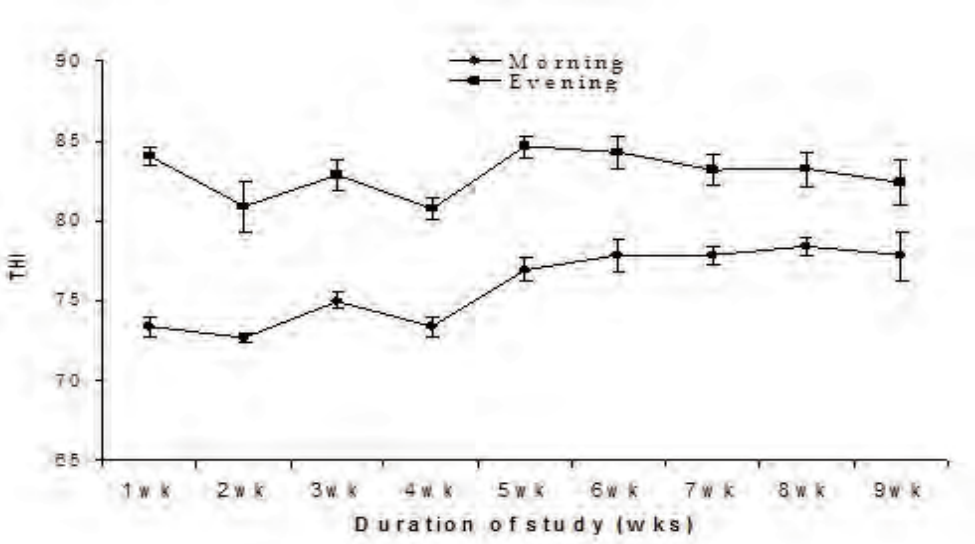

Fig 1. Graphical representation of changes in the temperature humidity index (THI) of morning and evening during the study periods (weekly intervals).

weeks was 73.35±0.69 and 84.01±0.62 which was change to 77.84±1.53 and 82.39±1.43 in the last weeks respectively. Significant increase ($p<0.05$) in catalase activity in group II as compared to other two other two groups at 4th wks of age however, no-significant change in the catalase activity was observed as the age progress in Vanaraja chickens. SOD activity significantly increase ($p<0.05$) in group II as compare to group I and II at 4th wks of age however, the significant increase ($p<0.05$) in activity was observed at 8th wks of age as compared to 4th wks of age. GSH-Px activity significantly lower ($p<0.05$) in the group II as compared to other groups however, the activity increases significantly ($p<0.05$) as the age progress i.e. at 8th wks as compared to 4th wks. Blood glutathione level was found maximum in group I as compared to group II and III and significantly lower in group II compared to group III at 4th wks of age of chicken. Whereas the blood level of GSH group III was significantly higher then the other two groups at 8th wks of age. The values increase significantly higher at 8th wks as compared to 4th wks of age of Vanaraja chicken. Non-significant changes were observed in hemoglobin concentration during the study both between age groups as well as the age progresses. To access stress induced cellular damage by the estimating the levels of peroxidation of lipid membrane. Lipid peroxidation significantly higher in group I as compared to group II and III at 4th wks of age and similar trend was also observed at 8th wks of age and as the age progress the lipid peroxidation increases.

Discussion

Mammalian cells are endowed with extensive antioxidant defense mechanisms which counteract the damaging effects of free radicals produced during the stress (Halliwell and Gutteridge, 1989). During stress, excess production of superoxide radicals in mitochondrial are converted to hydrogen peroxide and water with the help SOD (Andreyev *et al*., 2005). In present study, SOD activity significantly high in deep liter system as compared to semi-intensive and cage system suggests there was excess production of superoxide radicals. As the age progresses the production of superoxide radicals increases significantly, similar increased superoxide radical concentration with age has also been reported from different species including humans (Sohal *et al*., 2002, Valko *et al*., 2007). These superoxide radicals are metabolized to molecular oxygen and water by the catalase and in present study the activity of CAT increases significantly in deep liter system may be due to combat the excess production of superoxide radicals (Rayman, 2000). GSH-Px catalyzes reaction between inorganic or organic peroxides and reduced glutathione to form oxidized glutathione and water thus reduces the peroxides damaging effect (Rayman, 2000). In present study significantly reduced activity in deep liter system suggests that ability of the cell to remove/scavenge the excess of peroxides generated due to stress is reduced. GSH produces antioxidant potential in several ways, as a substrate for GSH-Px, well as a direct antioxidant thus protecting the cell from free radical damage. Significant decrease

in GSH level in deep liter system in present study may indicate either inhibition of GSH synthesis or increased utilization of GSH for scavenging the increased free radicals production (Singh *et al*., 2001). Heat-induced reactive oxygen species formation may be the factor that causes molecular changes in DNA, proteins, lipids and other biological molecules (Bruskov et al., 2002). Stress induced cellular damage was indicated by changes in the lipid peroxidation, and it was significantly higher in cage system at 4th and 8th wks of age, and increased significantly as the age progresses (Sen et al., 2006). Various other studies also suggested that the cage is more modern, beneficial and economic (Mohanlal, 1985). North (1984) estimated that 75% of all the commercial layers in the world are kept in cages and in the United States 93% of layers are in cages. Observations of the present study like increased blood level of GSH, higher activity of GSH-Px, comparable activity of CAT and SOD in cage system as compared to deep litter and semi-intensive system, suggested that cage system is better in handling the stress induced by different environmental factors.

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