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EFFECTS OF HOUSING SYSTEMS ON ANTIOXIDANTS STATUS OF LAYING HENS

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Abstract
The objective of the study was to determine the effect of housing system on the antioxidants status of laying hens. A total of 1000 laying hens were used for the study. One group was housed in deep litter while battery cage was used for the second group. Blood samples were collected from the wing vein at the age of 36 weeks and used for the determination of antioxidant status. Serum levels of superoxide dismutase (12.57±0.73 iu/L and 11.97±0.95 iu/L) and glutathione peroxidase (24.10±1.04 iu/L and 22.90±1.40 iu/L) were similar (P>0.05) for all the treatment groups. However, the serum level of catalase enzyme (36.43±1.22 iu/L and 31.50±1.61 iu/L) was significantly higher in birds on deep litter housing system. The serum level of malondialdehyde was significantly higher (P<0.05) in laying hens kept in battery cage (2.43±0.15 mmol/L) compared to (2.07±0.09 mmol/L) recorded in the deep litter housing system. The result indicated higher antioxidant capacity (2.27±0.20 and 1.63±0.12) in deep litter system while total oxidative stress (8.53±0.45 and 8.80±0.21) was similar for both housing systems. The oxidative stress index was significantly higher in the battery cage system (5.40±0.40) compared to (3.76±0.40) recorded in the deep litter housing system. It could be concluded therefore that laying hens kept in battery cages are more susceptible to oxidative stress compared to those housed in deep litter system
Keywords: Housing, layers, reactive oxygen species, oxidative stress

Introduction

Innate behaviour of laying hens including dust bathing, perching and nesting are internally and physiologically regulated. Depression arises when animals are inhibited from expressing naturally motivated behaviour (Frazer *et al*., 2013). The ability of hens to express physiologically motivated behaviour is central to achieving a positive welfare state (Meller and Webster, 2014).Although conventional cages contribute to prevention of infectious disease , ensure better hygiene and ease of management, hens experience extreme behavioural restrictions and lack of movement that predisposes them to metabolic disorders (Duncan, 2001).

Oxidative stress in poultry result from environmental, nutritional and management factors leading to adverse effects on poultry health and productivity. Oxidative stress result from in balance between free radical production and endogenous anti-oxidant defence leading to lipid peroxidation, protein oxidation and damage to DNA (Estevez, 2015). Reactive oxygen species are detoxified by superoxide dismuutase, catalase and glutathione peroxidase. Superoxide dismutase catalyse dismutation of superoxide to a normal oxygen molecule and hydrogen peroxide. The catalase enzyme decomposes hydrogen peroxide to water and hydroxyl radical, while glutathione peroxidase reduces lipid peroxidation (Fukai and Ushio, 2011).

Stress of any origin can induce oxidative stress by depleting antioxidant resources in the body, oxidative stress biomarkers are therefore reliable indicators of animal welfare status (Scanberg *et al*., 1993). High population density and poor housing conditions have been associated with increase oxidative stress in farm animals. The objective of this study was to determine the effects of housing systems on antioxidant status of laying hens.

Materials and Methods

Location of the study

The study was conducted at Ocean Blue Farms located in Kokona local government area of Nasarawa State. The farm is located on latitude 8.851097^0 and· longitude 7.9499^0. The area has a mean monthly temperature of 30 degree centigrade and annual rainfall of 1450mm (NIMET, 2008).

Experimental Birds and Management

A total of 1000 ISA Brown layers were used for the experiment. All recommended vaccinations were administered as required. All the birds were raised on deep litter system and at 16 weeks of age, 500 birds were moved to battery cages. The deep litter and battery cage systems served as the treatments with (500 birds each). Apart from the housing systems, all other management practices were same for all treatments. At 38 weeks of age, 3 mls of Blood samples were collected through the wing vein and prepared for analysis of antioxidants status following standard procedure.

Determination of Antioxidant Status

Serum level of superoxide dismutase was determined by calorimetric method based on the ability of the enzyme to inhibit phenazine methosulphate mediated reduction of nitro blue as described by Weydert (2003). Catalase activity was measured using combination of optimized enzymatic conditions and spectrophotometric assay of hydrogen peroxide as described by Laszo *et al*. (1991). Glutathione peroxidase activity was measured using hydrogen peroxide as a substrate as described by Liu, (2004). Serum malondialdehyde was determined in terms of thiobarbituric acid reactive substances using spectrophotometer as reported by Surapon, (2009). Total antioxidant capacity was determined by calorimetric method as described by Rahnama *et al*. (2015). Total oxidative stress was determined by calorimetric method based on the oxidation of ferrous ion to ferric ion in the presence of various oxidants in acidic medium as described by Ozcan, (2005).

Statistical Analysis

Data collected was subjected to analysis of variance (ANOVA) using SPSS (21.0) computer software. Significant differences between treatment means were separated using the Duncan Multiple Range Test. (SPSS, 2010).

Results and Discussion

The effect of housing systems on serum level of antioxidant enzyme is shown in table 1.

Table 1: Effects of Housing System on Serum Level of Antioxidant Enzymes (Mean±SEM)

Parameters	Determined Deep litter	Standard values with reference	Determined Battery cage	Standard values with reference	LOS
Superoxide dismutase (iu/L)	12.57±0.73	9.0 iu/L Muchaka *et al.* (2018)	11.97±0.95	11.0 iu/L Muchaka *et al*. (2018)	NS
Catalase (iu/L)	36.43±1.22[a]	31.0 iu/L Muchaka *et al*. (2018)	31.50±1.61[b]	17.0 iu/L Muchaka *et al*. (2018)	*
Glutathione peroxidase (iu/L)	24.10±1.04	21.6 iu/L Simek *et al*. (2015)	22.90±1.40	26.7 iu/L Simek *et al*. (2015)	NS
Malondialdehyde (mmol/L)	2.07±0.09[a]	1.92 mmol/L Simek *et al*. (2015)	2.43±0.15[b]	2.33 mmol/L Simek *et al*. (2015)	*

Means within same row bearing different superscript are significantly different (P<0.05), SEM – Standard Error or mean, Ns = Not significant, * = Significant (P<0.05), Los = Level of Significance

The concentrations of superoxide dismutase and glutathione peroxidase were similar (P>0.05) for both groups. Catalase activity was significantly higher (P<0.05) in birds housed in deep litter system (36.43±1.22), whereas the serum level of malondialdehyde was significantly higher (P<0.05) in the battery cage system (2.43±0.15). Similar studies indicated significantly higher serum levels of malondialdehyde in cage housing compared to floor system. The levels of gluthation peroxidase and catalase were however, similar for both groups Simsek *et al*. (2015). Significant decrease in superoxide dismutase activity was observed in cage system compared to floor system, while gluthation peroxidase activity was not affected by housing system Mamoud *et al*. (2020). Similarly, non-significant effect of housing system on superoxide dismutase, catalase and gluthatione peroxidase activities was documented by Mamoud and Mohammed, (2020). Variations in antioxidant status of layer chickens in different locations may be attributed to differences in environmental conditions. High ambient temperature is reported to modulate the expression of antioxidant enzyme genes which in turn regulate the synthesis of antioxidant enzymes Goel *et al*. (2021).
The effect of housing system on antioxidant status of laying hens is shown in table 2. The result indicated higher level (P<0.05) of total antioxidant capacity in the deep litter system (2.27±0.20). Total oxidative stress was similar for both groups (8.53±0.45 - 8.80±0.21), whereas, the oxidative stress index was higher in battery cage system (5.40±0.40).

Table 2: Effects of Housing System on Antioxidant Status of Laying Hens (Mean±SEM)

Parameters	Determined Deep litter	Standard values with reference	Determined Battery cage	Standard values with reference	LOS
Total antioxidant capacity	2.27±0.20[a]	2.46 Fathi *et al*. (2019)	1.63±0.12[b]	2.0 Li *et al*. (2019)	*
Total oxidative stress	8.53±0.45		8.80±0.21		NS
Oxidative stress index	3.76±0.40[a]		5.40±0.40 [b]		

Means within same row bearing different superscript are significantly different (P<0.05), SEM – Standard Error or mean, Ns = Not significant, * = Significant (P<0.05), Los = Level of Significance

A balance in cellular reactive oxygen species is modulated by processes that produces free radicals and the antioxidant system. Reactive oxygen species are mostly produced as products of oxidative metabolism and cellular responses to xenobiotics, cytokines and bacterial invasion (Galluzzi *et al*., 2010). Cellular oxidation produces superoxide which is detoxified by superoxide dismutase, a process that yield hydrogen peroxide as a by-product. Hydrogen peroxide is reduced to water by catalase and glutathione peroxidase enzymes (Turrens, 2003). The effects of reactive oxygen species on proteins, lipids and DNA are dependent on their concentrations, increased levels are present during oxidative stress (Christin and Joseph, 2010). High concentration of reactive oxygen species is cytotoxic, while lower levels play critical roles in several physiological processes including cell differentiation, apoptosis and regulation of redox sensitive signals. Oxidative stress induced damage can lead to cell death, mutation and cancer. The cellular concentration of reactive oxygen species is determined by rate of production and removal by the antioxidant system. The primary antioxidant enzymes in mammalian cells are superoxidedismutase, catalase and glutathione peroxidase (Liu, 2004). The concentration of malondialdehyde is an established indicator of cellular injury as well as a biomarker of oxidative stress in cells and tissues (Simek *et al*., 2006).
Animal welfare involves a combination of adequate nutrition, appropriate environment, optimum health and expression of normal behaviour. Conventional poultry cages lack adequate space resulting in behavioural restrictions and associated metabolic diseases (Hurtches and Jones, 2019). Dietary supplementation with vitamins C and E was reported to improve antioxidant and immune status. Dietary curcumin supplementation resulted in

improved antioxidant status and amelioration of stressful environmental conditions in laying hens (Aamir *et al*., 2019). The beneficial effects of ginger powder in improving antioxidant enzymes activity in serum and egg yolk of laying hens have been documented (Zhao *et al*., 2011). It can be concluded that the hens in battery cage system may be associated with reduced total antioxidant capacity and increased oxidative stress index. The higher serum level of malondialdehyde in the battery cage system is an indication of increased lipid peroxidation. Regular supplementation with antioxidants is recommended in the battery cage system of poultry production to ameliorate the effects of stress.

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